VOLUME 81 *February 1960* NUMBER 2

THE AMERICAN REVIEW OF RESPIRATORY DISEASES

Clinical and Laboratory Studies of Tuberculosis and Respiratory Diseases

OFFICIAL JOURNAL OF AMERICAN TRUDEAU SOCIETY
MEDICAL SECTION OF THE NATIONAL TUBERCULOSIS ASSOCIATION

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(Formerly THE AMERICAN REVIEW OF TUBERCULOSIS AND PULMONARY DISEASES)

ANNUAL MEETING, LOS ANGELES, CALIFORNIA, May 15–20, 1960

See page 4 of advertisements

Published Monthly at Mount Royal and Guilford Avenues, Baltimore 2, Maryland, by the National Tuberculosis Association. Business Off., 1790 Broadway, New York 19, N. Y.

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THE AMERICAN REVIEW OF RESPIRATORY DISEASES

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Membership: All physicians and surgeons in the United States, Canada, and other countries who are interested in tuberculosis and other respiratory diseases may apply for membership in the American Trudeau Society. The dues for active membership in the Society are $18 per year, which includes membership in the National Tuberculosis Association, copies of the Society's News Letter, and a subscription to the REVIEW and to the monthly BULLETIN of the National Tuberculosis Association.

Subscriptions: The subscription price of the REVIEW is $15 for the calendar year. Checks should be made payable to the National Tuberculosis Association. The price of single copies is $2.00 postpaid.

Character of the Review: The REVIEW invites the submission of original manuscripts on any phase of tuberculosis or respiratory diseases.

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Contributors are requested to pay particular attention to the following rules governing the preparation of manuscripts. Failure to follow these rules is a frequent cause of delay in the publication of articles.

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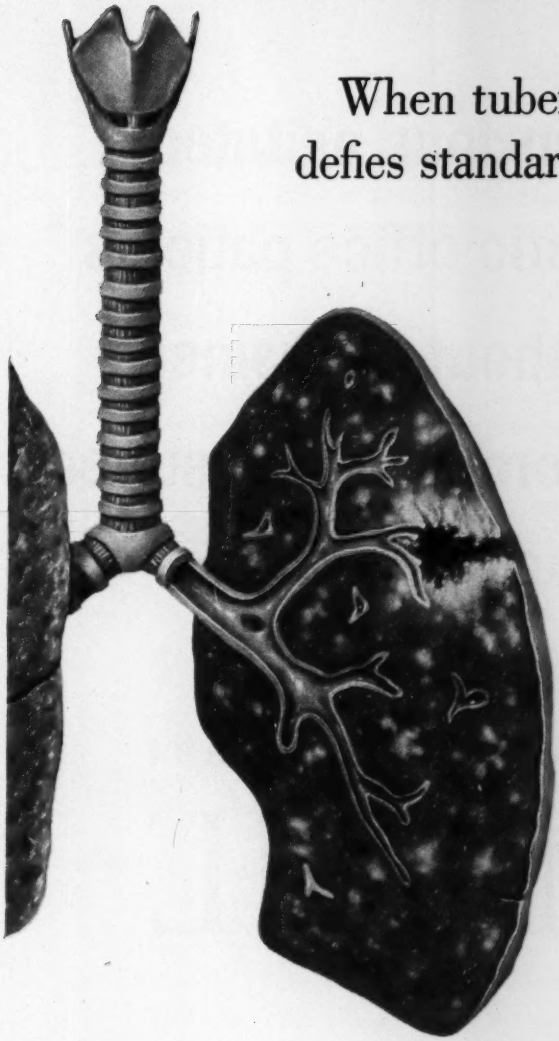

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Registration will be in the Statler Hilton and will begin at 9:00 AM, SUNDAY, MAY 15.

Exhibits will be in the Statler Hilton and will open for viewing at 8:30 AM, MONDAY, MAY 16.

Special interest sessions will begin SUNDAY, MAY 15. *Medical, public health, and nursing sessions* will begin MONDAY, MAY 16.

General sessions will include an opening keynote program Monday at 9:00 AM, followed immediately by the awarding of the Trudeau and Will Ross medals; a session on Monday afternoon at which results of the Arden House Conference on Tuberculosis will be presented (the opening portion will be of special interest to doctors); a session Tuesday morning, and the NTA Business Luncheon on Thursday.

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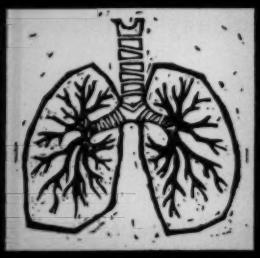

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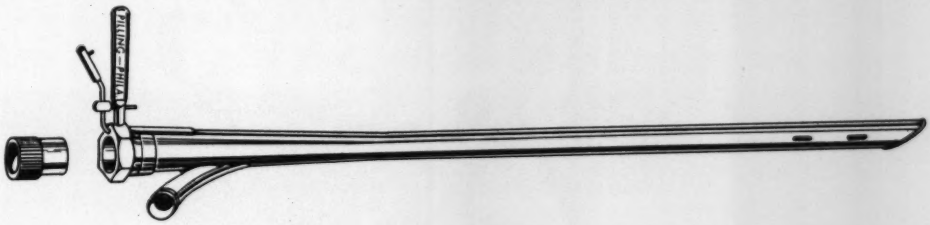

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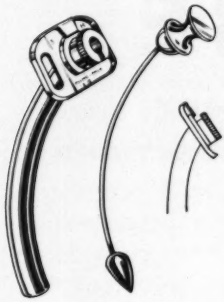

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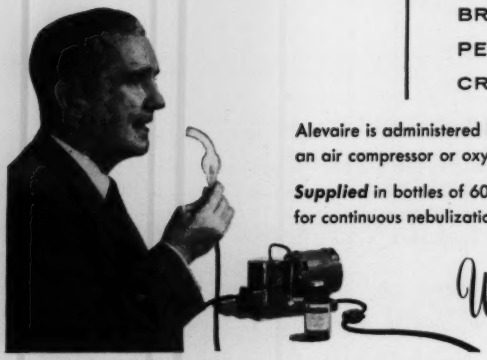

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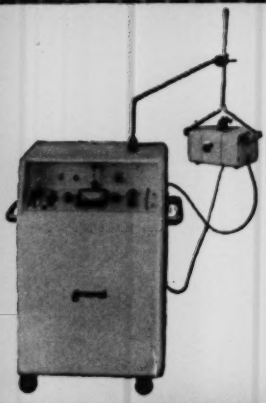

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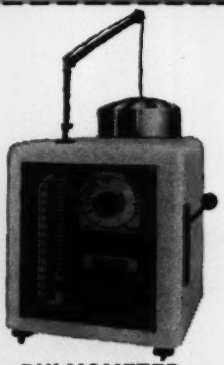

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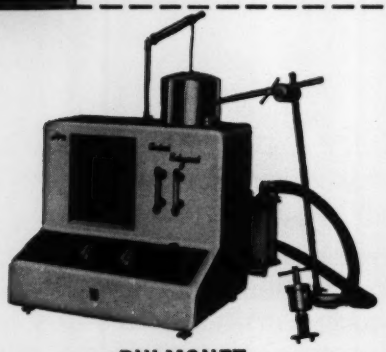

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Clinical and Laboratory Studies of Tuberculosis and Respiratory Diseases

VOLUME 81 *February 1960* NUMBER 2

A STUDY OF FIVE HUNDRED CONSECUTIVE ADMISSIONS TO A TUBERCULOSIS HOSPITAL[1, 2]

C. A. WICKS

(Received for publication July 6, 1959)

Introduction

A study of consecutive admissions to a hospital has the advantage of revealing a true unselected sampling or cross section of the type of patients admitted, the nature and extent of their illnesses, the treatments applied, as well as the progress observed. Entrants to a tuberculosis hospital, particularly those patients with pulmonary tuberculosis, lend themselves to such a study because of reasonably well-defined categories or classifications according to extent of disease, presence or absence of cavitation, and bacteriologic findings. Conferences, e.g., the Pembine type of conference, have been developed to report upon consecutive hospital admissions, and the case of each patient is presented for critical analysis. The method of reporting each case in detail at such conferences has usually restricted the number of consecutive admissions that could be considered. This paper will report statistically upon 500 consecutive admissions to the Toronto Hospital for Tuberculosis at Weston from January 1 to August 12 (inclusive), 1957.

During the period of this study there was no special section for nontuberculous patients in the Toronto Hospital for Tuberculosis. All of the patients entered for the review or treatment of tuberculous disease or for the investigation of suspected tuberculosis.

Among the 500 consecutive entries considered, 3 patients had more than one entry. Each of these 3 patients entered twice during the period under study. The second entries were excluded, leaving 497 admissions for statistical analysis.

Method

The medical records of the 497 patients were reviewed in detail. For each patient a specially designed card was completed, recording the required information opposite suitable headings. No attempt was made to extend the information beyond what was available on file. Copies of reports upon the examination of discharged patients are filed as completed or as received from chest clinics and other sanatoriums. The information contained in the medical records was accepted and used in recording the condition of each patient on entry and at intervals of three months up to eighteen months after entry. The cards were then sorted according to selected variables.

The following definitions are employed:

Readmission refers to a patient previously in any sanatorium and who is readmitted for treatment after being regularly discharged, or who re-enters the sanatorium for treatment more than thirty days after premature discharge. (Patients who re-enter the same sanatorium within thirty days after leaving without medical consent are not classified as premature discharges, but are considered merely as returned from leave.)

Transfer in refers to a patient who is transferred directly from another sanatorium or who enters within thirty days after leaving another sanatorium prematurely.

For review refers to a readmission, usually for reassessment when extended sanatorium care is not required.

Familial descent refers to the ethnic group from which a person is descended. For statistical purposes, origin is determined through the father.

Previous sanatorium treatment adequate is considered to have been received if the

[1] From the Toronto Hospital for Tuberculosis, Weston, Ontario, Canada.

[2] Presented in part at the annual meeting of the Canadian Tuberculosis Association, Halifax, Nova Scotia, June, 1959.

patient was last discharged from a sanatorium with medical consent and had received a total of at least twelve consecutive months of chemotherapy (including any post-sanatorium chemotherapy), using concurrently at least two of the drugs, streptomycin, PAS, or isoniazid.

Bacteriologic confirmation refers to a patient from whom one or more specimens of sputum or pleural fluid revealed acid-fast bacilli on smear or organisms indistinguishable from tubercle bacilli on culture within three months before, or three months after, entry (also gastric washings, but on culture only).

Drug resistance—any degree refers to the results of drug-susceptibility tests performed by the technique in use at the Toronto Hospital for Tuberculosis, using subcultures on Herrold's egg yolk agar containing glycerol of selected colonies previously cultured on a slope of modified Löwenstein medium:

To streptomycin—any growth in the presence of 10 γ of streptomycin per ml. of medium

To PAS—any growth in the presence of 10 γ of PAS per ml. of medium

To isoniazid—any growth in the presence of 1.0 γ of isoniazid per ml. of medium

Drug resistance—definite refers to the finding of growth on the plate with the above-mentioned concentration for the drug in question, equaling the growth on the control plate containing no drug.

Previous chemotherapy refers to previous treatment with the drug as stated (if not stated, one or more of streptomycin, PAS, or isoniazid) for a continuous period of more than one month at any time prior to admission.

Result satisfactory refers to the status of a patient with pulmonary tuberculosis without evidence of cavitation (by planigraphy if cavity was previously shown by this method) whose secretions are negative for tubercle bacilli on microscopy and culture and who shows stable or unchanging roentgenographic findings.

Unsatisfactory progress includes those who died from tuberculosis, those who remained in the sanatorium eighteen months after entry, and those who had not achieved satisfactory results when regularly discharged.

Satisfactory progress signifies achievement of satisfactory results before or at regular discharge within eighteen months after entry.

Interruption of streptomycin, PAS, and isoniazid was considered to be the omission of one or more of those drugs for a consecutive period of more than one month.

Results

Distribution by age and sex groups: There were 281 male and 216 female patients comprising the group studied (table 1). The 497 patients were grouped into 350 first admissions, 84 readmissions, 39 entering as transfers from another hospital, and 24 for review of disease status.

The peak of male and female entries occurred in the twenty- to twenty-nine-year age group. This peak was higher for female than for male patients, but the excess of male patients over female patients among entrants more than fifty years old is striking. It is worthy of note that 29.2 per cent of those entering the hospital were fifty years of age or older.

Familial descent: English, Irish, and Scottish parentage accounted for 51.9 per cent of all persons admitted to the hospital. The period under study coincided with the immigration of a number of persons from Hungary without the advantage of screening before leaving Europe. Hungarians, therefore, accounted for 6.6 per cent of all entries. No Eskimo patients were among the group. Only 12 (2.4 per cent) were North American Indians, and 7 of that number entered by transfer from another hospital.

Country of birth and years in Canada: Persons born in Canada accounted for 55.3 per cent of

TABLE 1
Age and Sex Groups

	Totals				
	Male	Female	Total	Per Cent	
Totals	281	216	497	100	
years					
0 to 9	4	4	8	1.6	
10 to 19	18	21	39	7.8	
20 to 29	60	68	128	25.8	
30 to 39	47	62	109	21.9	
40 to 49	35	33	68	13.7	
50 to 59	56	9	65	13.1	
60 to 69	35	9	44	8.9	29.2%
70 to 79	24	8	32	6.4	
80 and Older	2	2	4	0.8	
First admission	—	—	350	70.4	
Readmission	—	—	84	17.0	
Transfer in	—	—	39	7.8	
For review	—	—	24	4.8	

TABLE 2

TYPE AND EXTENT OF DISEASE

	Totals			First Admissions		Readmissions	Transfer in	For Review
	Number	Per Cent of Entries	Per Cent of Group	Number	Per Cent			
Totals	497	100	—	350	100	84	39	24
Respiratory tuberculosis	405	81.5	100	284	81.1	69	36	16
(with other tuberculosis)	31	6.2	7.7	21	6.0	6	2	2
Primary type	8	1.6	2.0	7	2.0	1	0	0
Pleurisy with effusion	14	2.8	3.5	13	3.7	0	1	0
Minimal	135	27.2	33.3	112	32.0	9	10	4
Moderately advanced	139	28.0	34.3	91	26.0	29	12	7
Far advanced	100	20.1	24.7	54	15.4	29	12	5
Unclassified with silicosis	9	1.8	2.2	7	2.0	1	1	0
Nonrespiratory tuberculosis	66	13.3	100	41	11.7	15	3	7
(with other tuberculosis)	32	6.4	48.5	16	4.6	8	1	7
Genitourinary	23	4.6	34.8	12	3.4	3	2	6
Bone and joint	16	3.2	24.2	7	2.0	7	1	1
Pelvic (female)	15	3.0	22.7	13	3.7	2	0	0
Lymph nodes	8	1.6	12.1	7	2.0	1	0	0
Others	4	0.8	6.1	2	0.6	2	0	0
Nontuberculous disease only	26	5.2	100	25	7.1	0	0	1
Respiratory—malignant	11	2.2	42.3	11	3.1	0	0	0
Respiratory—other	6	1.2	23.1	5	1.4	0	0	1
Genitourinary	4	0.8	15.4	4	1.1	0	0	0
Cardiovascular	2	0.4	7.7	2	0.6	0	0	0
Lymph nodes	1	—	3.8	1	—	0	0	0
Bone and joint	1	—	3.8	1	—	0	0	0
Pelvic (female)	1	—	3.8	1	—	0	0	0

all entries; 15.5 per cent of the group were born in the United Kingdom; 6.2 per cent, in Hungary; fewer than 1 per cent had been born in the United States.

Of the 222 entrants not born in Canada, 44.3 per cent had lived in Canada less than six years. The number of new Canadians in this group who entered the Weston sanatorium decreased as the number of years of their stay in Canada increased.

Type and extent of disease: It may be seen in table 2 that, among the 497 entries studied, 405 (81.5 per cent) of the patients had tuberculosis of the respiratory system as the most important problem, 66 (13.3 per cent) had other forms of tuberculosis predominantly, while 26 (5.2 per cent) had only nontuberculous disease.

Patients with respiratory tuberculosis as the principal problem had recorded incidences of other tuberculous disease (active or inactive) in 7.7 per cent of cases, whereas, for patients with nonrespiratory tuberculosis as the main problem, the comparable figure was 48.5 per cent.

Among the patients with respiratory tuberculosis as the principal problem, 2.0 per cent had the primary type; 3.5 per cent, pleurisy with effusion; 33.3 per cent, minimal disease; 34.3 per cent, moderately advanced disease; 24.7 per cent, far advanced disease; and 2.2 per cent, unclassified pulmonary tuberculosis with silicosis.

Among the patients with nonrespiratory tuberculosis as the main problem, the majority (34.8 per cent) had genitourinary tuberculosis; 24.2 per cent had bone and joint tuberculosis; 22.7 per cent had tuberculosis of the female reproductive system; 12.1 per cent had tuberculosis of superficial lymph nodes.

Among the 26 persons who were found after entry to have only nontuberculous disease, cancer of the lung was most common (11 patients) and

TABLE 3

REASON FOR EXAMINATION LEADING TO ENTRY

(First Admissions and Readmissions with Active Respiratory Tuberculosis)

	Total		First Admissions		Readmissions	
	Number	Per Cent	Number	Per Cent	Number	Per Cent
Active respiratory tuberculosis	350	100	281	100	69	100
Symptoms—Total	143	40.9	110	39.1	33	47.8
Routine examinations—Total	207	59.1	171	60.9	36	52.2
Employment	76	21.7	71	25.3	5	7.2
Other "survey"	33	9.4	32	11.4	1	1.4
Post-sanatorium follow-up study	25	7.1	—	—	25	36.2
Hospital admission	22	6.3	21	7.5	1	1.4
Contact	22	6.3	22	7.8	0	—
Immigration	16	4.6	13	4.6	3	4.3
School or college	8	2.3	8	2.8	0	—
Jail	5	1.4	4	1.4	1	1.4
Last previous hospital treatment adequate	8	—	—	—	8	11.6

other nontuberculous respiratory disease was the next most common finding (6 patients).

Commonly associated nontuberculous conditions on hospital entry: The prevalence of nontuberculous conditions associated with tuberculosis was determined as diagnosed and recorded on the patients' charts by the staff physicians. Among 471 entrants with tuberculosis, chronic alcoholism in 20 patients (4.2 per cent) was recorded as an associated diagnosis. Eighteen patients (3.8 per cent) had associated nontuberculous respiratory disease; 17 (3.6 per cent) had previously had thoracoplasties; 14 (3 per cent) had chronic cardiovascular disease; 13 (2.8 per cent) had diagnoses of associated psychoneurosis or mental illness; 12 (2.5 per cent) had associated diabetes mellitus; 8 women (4 per cent of female entries with tuberculosis) were pregnant on entry; 7 (1.5 per cent) had nonrespiratory malignant disease; 5 (1.1 per cent) had partial or complete gastrectomies performed at some time prior to admission; and 6 patients (1.3 per cent) had had pulmonary resections previously performed. The last-named group consisted of 3 patients who were admitted merely for review and 3 who were transferred from other hospitals, one of whom had just had a pulmonary resection.

Reason for examination leading to entry: From the total of 497 patients admitted, 147 were excluded from analysis (table 3). The group excluded consisted of: 36 patients transferred from other hospitals; 16 admitted solely for review; 66 with nonrespiratory tuberculosis; 3 with clearly inactive tuberculosis; and 26 patients who were found to have no tuberculosis. Thus, there remained 350 patients with active respiratory tuberculosis for more detailed study.

Symptoms of ill health caused the patient to consult a physician, chest clinic, or chest roentgenographic screening station for an examination leading to a diagnosis of active respiratory tuberculosis in 39.1 per cent of 281 first admissions and 47.8 per cent of the 69 readmissions. Thus, the importance of the personal physician or the referral type of chest clinic in the detection of respiratory tuberculosis should not be overlooked, except that it would be desirable to detect active tuberculous disease before it gives rise to symptoms. Although not shown in the table, among the patients whose disease was detected because of symptoms, 39 per cent had far advanced pulmonary tuberculosis whereas, among those detected by routine examinations, only 13 per cent had far advanced pulmonary tuberculosis. Routine roentgenographic examination of the chest in association with employment (before, on, or during employment) resulted in the detection of active respiratory tuberculosis in 25.3 per cent of the first admissions and in 7.2 per cent of the readmissions. The examination of tuberculosis contacts, although accounting for only 7.8 per cent of first admissions, did yield

TABLE 4

CONDITION ON ENTRY

(First Admissions and Readmissions with Active Respiratory Tuberculosis)

	First Admissions			Readmissions		
	Number	Per Cent of First Admissions	Per Cent of Tested	Number	Per Cent of Readmissions	Per Cent of Tested
Totals	281	100		69	100	
Cavity definite	69	24.6		22	31.9	
Started on streptomycin + PAS + isoniazid	261	92.9		57	82.6	
PAS + hydrazone of isoniazid	12	4.3		0	0	
Other 2 drug regimens	6	2.1		9	13.0	
None or 1 drug only	2	0.7		3	4.4	
Bacteriologic confirmation	201	71.5		51	73.9	
Culture positive for tubercle bacilli—entry specimen	177	63.0		42	60.9	
Tested for susceptibility to streptomycin	166	59.1		38	55.1	
Resistance—any degree	41	14.6		11	15.9	
—definite	13	4.6	7.8	7	10.1	18.4
Previous chemotherapy with streptomycin	1	0.4		9	13.0	
No resistance of any degree	125	44.5		27	39.2	
Previous chemotherapy with streptomycin	0	0		10	14.5	
Tested for susceptibility to PAS	166	59.1		37	53.6	
Resistance—any degree	5	1.8		5	7.2	
—definite	1	0.4	0.6	3	4.3	8.1
Previous chemotherapy with PAS	0	0		5	7.2	
No resistance of any degree	161	57.3		32	46.4	
Previous chemotherapy with PAS	0	0		12	17.4	
Tested for susceptibility to isoniazid	165	58.7		37	53.6	
Resistance—any degree	10	3.6		11	15.9	
—definite	5	1.8	3.0	8	11.6	21.6
Previous chemotherapy with isoniazid	1	0.4		9	13.0	
No resistance of any degree	155	55.1		26	37.7	
Previous chemotherapy with isoniazid	0	0		6	8.7	
Tested for susceptibility to streptomycin + PAS + isoniazid	165	58.7		36	52.2	
Resistance—*any degree*—1 drug only	37	13.2		9	13.0	
—2 or more	9	3.2		7	10.1	
—1 or more	46	16.4		16	23.1	
Resistance—*definite*—1 drug only	15	5.3		5	7.2	
—2 or more	2	0.8		5	7.2	
—1 or more	17	6.1	10.3	10	14.4	27.8

a relatively large number of "early" cases. (Although not shown in the table, only 2 of 22 patients detected by this method had far advanced pulmonary tuberculosis.) The routine follow-up examinations of ex-sanatorium patients accounted for 36.2 per cent of readmissions with active respiratory tuberculosis.

It is noteworthy that, among this group, only

8 (11.6 per cent) of 69 readmissions were considered to have had relapse of their disease after a previous period of adequate hospital treatment, as previously defined.

Condition on entry: In table 4 may be seen the condition on entry of the 350 patients (281 first examinations and 69 re-examinations) with active respiratory tuberculosis. Definite cavitation was present in 24.6 per cent of first admissions and in 31.9 per cent of readmissions. Triple-drug therapy, using streptomycin, PAS, and isoniazid concurrently, was prescribed and was started for 92.9 per cent of first admissions and for 82.6 per cent of readmissions. There was bacteriologic confirmation of the diagnosis of tuberculosis in 71.5 per cent of first admissions and in 73.9 per cent of readmissions. Tubercle bacilli were isolated from the specimens obtained on admission to the hospital in 63.0 per cent of the first admissions and in 60.9 per cent of the patients who represented readmissions.

Tubercle bacilli with definite resistance to streptomycin were isolated on entry from 13 (4.6 per cent) of all first admissions or from 7.8 per cent of first admissions tested. All but one of these patients claimed to have had no previous treatment with streptomycin. Strains with definite resistance to streptomycin were likewise found in 10.1 per cent of all readmissions or in 18.4 per cent of readmissions tested. All readmissions whose strains of tubercle bacilli showed definite resistance to streptomycin had received previous treatment with streptomycin.

Tubercle bacilli with definite resistance to PAS were found on entry in one (0.4 per cent) of all first admissions, or 0.6 per cent of first admissions tested. (This one patient claimed to have had no previous treatment with PAS.) The comparable figures for readmissions were 4.3 and 8.1 per cent. All readmitted patients with strains of tubercle bacilli definitely resistant to PAS had received previous treatment with PAS.

Tubercle bacilli with definite resistance to isoniazid were found on entry in 5 (1.8 per cent) of all first admissions or 3.0 per cent of first admissions tested. Four of the 5 patients claimed to have had no previous treatment with isoniazid. Strains with definite resistance to isoniazid were found in 11.6 per cent of all readmissions, or 21.6 per cent of readmissions tested. All patients readmitted who were discharging tubercle bacilli definitely resistant to isoniazid had received previous treatment with isoniazid.

In the last line of table 4, it may be seen that *tubercle bacilli definitely resistant to one or more of the three drugs* studied were isolated on entry from 6.1 per cent of all first admissions with active respiratory tuberculosis, or 10.3 per cent of such first admissions tested. The comparable figures for readmissions were 14.4 and 27.8 per cent.

TABLE 5

THERAPEUTIC PROGRESS DURING 18 MONTHS AFTER ENTRY
(First Admissions and Readmissions with Active Respiratory Tuberculosis)

	Number	Remarks
Total (A)	350	
Prematurely discharged (B)	49	14.0% of total
Transferred out (C)	16	4.6% of total
Deaths due to nontuberculous causes (D)	9	2.6% of total
Net total (E)	276	For assessment
Deaths due to tuberculosis (F)	8	2.9% of net total
Remaining in sanatorium at 18 months (G)	14	5.1% of net total
Result unsatisfactory at regular discharge (H)	17	6.1% of net total
Unsatisfactory progress (F + G + H) (I)	39	14.1% of net total
Satisfactory progress—result satisfactory before or at regular discharge (J)	237	85.9% of net total
Maintenance of satisfactory progress after regular discharge: Result satisfactory at 18 months after entry:		
Status of tuberculosis known	144	
Result satisfactory	144	100% of status known

It is evident that previous chemotherapy significantly increased the prospect that drug-resistant bacilli could be cultured.

Progress during eighteen months after entry: The progress of 350 patients with active respiratory tuberculosis on entry is presented in summary form in table 5. After deducting 49 patients prematurely discharged, 16 transferred, and 9 who died from nontuberculous diseases, a net total of 276 patients is left for assessment. Among the last-named group, there were 8 patients who died from tuberculosis, 14 who were still in the hospital at eighteen months after entry, and 17 who had not achieved satisfactory results when discharged. Thus, there was a total of 39 patients (14.1 per cent of the net total) who had what might be considered unsatisfactory progress. The remaining 237 patients (85.9 per cent of the net total) had what might be considered as satisfactory progress and achieved satisfactory results before or at regular discharge within eighteen months after entry.

It is interesting to note that information was available on the status of the tuberculosis at eighteen months after entry of 144 of the 237 patients who made satisfactory progress in the hospital and were discharged as having attained satisfactory results. At this eighteen-month observation point, the therapeutic result in all 144 patients could be classified as satisfactory. Therefore, as far as the information available on file was concerned, all patients in this group who were discharged as having attained "satisfactory results" had maintained their satisfactory progress and remained in that favorable condition eighteen months after entry. (This last statement has obvious limitations in view of the short period involved and the lack of any attempt to obtain follow-up information for those for whom such information was not on file.)

Nine (18.4 per cent) of the 49 patients who were discharged prematurely were infectious (sputum culture) at the last examination before leaving the hospital. Three of the 9 patients were discharging tubercle bacilli that, at the last examination, had shown some degree of resistance to one or more of the three drugs used. Among these 49 premature dischargees were 8 patients (16.3 per cent) with chronic alcoholism. These 8 patients represented 41.7 per cent of those with a diagnosis on entry of chronic alcoholism associated with active respiratory tuberculosis. The status of the tuberculous disease at eighteen months after entry was known for only 11 of the 49 premature dischargees. One had died from tuberculosis, 4 had returned and were in a tuberculosis hospital, while 6 could be classified as "result satisfactory." Thus, 5 of the 11 showed unsatisfactory progress with respect to their disease at the eighteen-month observation point.

The nontuberculous causes of death were: bronchogenic cancer, 1; cancer of rectum, 1; cancer of stomach, 1; cancer of mouth, 1; malignant cholangioma of liver, 1; cancer of prostate with uremia, 1; cancer of prostate with secondaries in lung, 1; myocardial infarction, 1; acute hemorrhagic pancreatitis, 1. These 9 patients had roentgenographic and/or bacteriologic evidence of active pulmonary tuberculosis on entry. The causes of death were established by postmortem examinations in 6 cases, histologic evidence in 2 cases, and electrocardiographic findings in one case.

Effect of drug resistance upon progress of disease: An attempt is made in table 6 to correlate drug resistance and the course of the tuberculosis while the patient was in the hospital. As drug-susceptibility studies could only be performed on patient-strains isolated on culture, the percentage (80.4 per cent) of the group with satisfactory progress who were discharging tubercle bacilli at the time of hospital entry may be used as a baseline for comparison.

Drug resistance of a patient-strain on entry *of any degree* to one drug only did not reduce the satisfactory progress rate, whereas drug resistance of any degree to two or more drugs did have a significantly unfavorable effect upon progress.

Definite resistance of a patient-strain on entry to one drug only appeared to have some unfavorable effect upon the course of disease. Definite resistance to two or more drugs had a markedly unfavorable effect upon progress.

Patients who had had no previous chemotherapy when admitted to the hospital, whether with or without drug-resistant strains on entry, showed no depression of satisfactory progress of their disease during therapy. Those with previous chemotherapy and no definite drug resistance on entry also showed the expected percentage with satisfactory progress; but those with previous chemotherapy plus drug-resistant strains of tubercle bacilli on entry showed unsatisfactory progress.

Reading horizontally in the last section of

TABLE 6

EFFECT OF DRUG RESISTANCE UPON THERAPEUTIC PROGRESS IN THE HOSPITAL

(First Admissions and Readmissions with Active Respiratory Tuberculosis Excluding Premature Discharges, Transfers out, and Nontuberculous Deaths)

	Net Total for Assessment	Unsatisfactory Progress	Satisfactory Progress	
			Number	Per Cent of Net Total
Total	276	39	237	85.9
Bacteriologic confirmation	199	39	160	80.4
Resistance on entry: any degree—1 drug only	36	7	29	80.6
—2 or more	14	7	7	50.0
—1 or more	50	14	36	72.0
Resistance on entry: definite—1 drug only	17	5	12	70.6
—2 or more	6	4	2	33.4
—1 or more	23*	9	14	60.9
No previous chemotherapy—no definite drug resistance on entry	227	25	202	89.0
—definite drug resistance—1 or more on entry	12*	2	10	83.3
Previous chemotherapy—no definite drug resistance on entry	26	5	21	80.8
—definite drug resistance—1 or more on entry	11*	7	4	36.4

* Type of definite drug resistance on entry

	No Previous Chemotherapy	Previous Chemotherapy				
Streptomycin only	9	2	11	2	9	81.8
Isoniazid only	3	3	6	3	3	50.0
Streptomycin + isoniazid	0	3	3	1	2	66.6
Streptomycin + PAS + isoniazid	0	3	3	3	0	0
Total	12	11	23	9	14	60.9

table 6, it appears that, with the methods of treatment in use at the Toronto Hospital for Tuberculosis, satisfactory progress among the group studied was not depressed by definite resistance to streptomycin alone; but was depressed by definite resistance to isoniazid alone, also by definite resistance to streptomycin and isoniazid together. Satisfactory progress was entirely lacking among patients whose strains of tubercle bacilli at the time of hospital entry were definitely resistant to all three drugs used.

Effect of certain other factors upon progress: Attempts to correlate various other factors with progress in the hospital are set forth in table 7. Among patients with definite cavitation on entry, 67.6 per cent had satisfactory progress, compared with 92.6 per cent for those without cavitation. Among those with bacteriologic confirmation, 80.4 per cent showed satisfactory progress, compared with 100 per cent for those without bacteriologic confirmation. Interruption of concurrent three-drug therapy with streptomycin, PAS, and isoniazid appeared to have a somewhat unfavorable effect upon progress. This observation cannot be accepted as evidence for the superior merit of triple-drug therapy because only a small number received any other regimen than concurrent triple-drug therapy, and because of the lack of randomization in the selection of patients for the type of chemotherapy.

Among the patients in this group, the prospect

TABLE 7

EFFECT OF CERTAIN OTHER FACTORS UPON THERAPEUTIC PROGRESS IN THE HOSPITAL

(First Admissions and Readmissions with Active Respiratory Tuberculosis Excluding Premature Discharges, Transfers out, and Nontuberculous Deaths)

	Net Total for Assessment	Unsatisfactory Progress	Satisfactory Progress	
			Number	Per Cent of Net Total
Totals	276	39	237	85.9
Cavity definite on entry	74	24	50	67.6
No cavity or uncertain on entry	202	15	187	92.6
Bacteriologic confirmation	199	39	160	80.4
No bacteriologic confirmation	77	0	77	100
Interruption of streptomycin + PAS + isoniazid	41	10	31	75.6
No interruption of streptomycin + PAS + isoniazid	214	25	189	88.3
Primary type—first admission	6	0	6	100
—readmission	1	0	1	100
Pleurisy with effusion—first admission	12	0	12	100
—readmission	0	0	0	—
Minimal—first admission	88	3	85	96.6
—readmission	7	1	6	85.7
Moderately advanced—first admission	78	5	73	93.6
—readmission	18	2	16	88.9
Far advanced—first admission	41	16	25	61.0
—readmission	20	12	8	40.0
Unclassified with silicosis—first admission	4	0	4	100
—readmission	1	0	1	100
Pulmonary resection performed	28	1	27	96.4
Thoracoplasty only performed	1	1	0	0

of satisfactory progress (prognosis) decreased proportionately as the extent of the pulmonary tuberculosis increased. Also, for each extent of disease, the progress was less satisfactory for patients who were readmissions than for first admissions. Representative figures for satisfactory progress by extent of disease are: minimal first admissions, 96.6 per cent, readmissions 85.7 per cent; far advanced first admissions 61.0 per cent, readmissions 40.0 per cent.

There were available for assessment 28 pulmonary resections performed during the eighteen months after entry for the 276 patients with active respiratory tuberculosis. The resections consisted of: 2 pneumonectomies, 11 lobectomies, 13 segmental resections, and 2 wedge resections. Of the 28 patients who had pulmonary resection as part of their treatment, 27 (96.4 per cent) showed satisfactory progress. The one patient classified as showing unsatisfactory progress with pulmonary resection was a 40-year-old chronic alcoholic female (*C.W.*) with three previous admissions to tuberculosis hospitals. A thoracoplasty had been performed during a previous admission. On this last occasion, lobectomy under the previous thoracoplasty was performed sixteen months after entry. She remained in the sanatorium eighteen months after entry and, therefore, was classified as showing unsatisfactory progress, although she was regularly discharged nineteen months after entry with the classification "result satisfactory."

The one patient with thoracoplasty classified as showing unsatisfactory progress was a 57-year-old male (*S.Z.*). He had received streptomycin–isoniazid at home for twelve months before his first admission. On admission he had scattered bilateral opacities with gross unilateral cavitation. Tubercle bacilli cultured from all entry specimens showed a definite degree of resistance to streptomycin and isoniazid. Seven months after entry he had persisting cavitation in the left upper lung and he continued to be infectious with tubercle bacilli definitely resistant to streptomycin and isoniazid. At that time a seven-rib two-stage thoracoplasty was performed.

He was regularly discharged sixteen months after entry, but sputum was then occasionally positive for tubercle bacilli so that he was classified as showing unsatisfactory progress.

Drug resistance and reversal of infectiousness: The rapid rate of reversal of infectiousness may be seen in table 8. Tubercle bacilli could be isolated from 62 per cent of the patients on hospital entry, from 15.2 per cent at three months, and from 6.2 per cent at six months after hospital entry. In 5 patients from whom tubercle bacilli were not isolated at hospital

TABLE 8

FOLLOW-THROUGH STUDY OF SPUTUM CONVERSION AND DRUG RESISTANCE

(First Admissions and Readmissions with Active Respiratory Tuberculosis Excluding Premature Discharges, Transfers out, and Nontuberculous Deaths)

	On Entry Negative on Culture	On Entry Positive on Culture	Total		Entry Resistance 1 or More Any Degree	Entry Resistance 1 or More Definite
			Number	Per Cent of Total		
On entry	105	171	276	100	50	23
Positive on culture on entry	—	171	171	62.0	50	23
Susceptibility tested to all 3 drugs	—	160	160	58.0	50	23
Resistance to 1 or more drugs—any degree	—	50	50	18.1	50	—
—definite	—	23	23	8.3	—	23
Remaining in hospital at 3 months after entry	102	166	268	97.1	50	23
Positive on culture at 3 months after entry	5	37	42	15.2	15	11
Susceptibility tested to all 3 drugs	4	24	28	10.1	10	7
Resistance to 1 or more drugs—any degree	4	10	14	5.1	8	6
—definite	4	5	9	3.3	5	5
Remaining in hospital at 6 months after entry	94	164	258	93.5	49	22
Positive on culture at 6 months after entry	1	16	17	6.2	11	9
Susceptibility tested to all 3 drugs	0	7	7	2.5	5	5
Resistance to 1 or more drugs—any degree	—	5	5	1.8	4	4
—definite	—	5	5	1.8	4	4
Remaining in hospital at 9 months after entry	68	150	218	79.0	43	19
Positive on culture at 9 months after entry	1	14	15	5.4	10	8
Susceptibility tested to all 3 drugs	0	8	8	2.9	6	6
Resistance to 1 or more drugs—any degree	—	7	7	2.5	6	6
—definite	—	5	5	1.8	5	5
Remaining in hospital at 12 months after entry	23	69	92	33.3	24	13
Positive on culture at 12 months after entry	0	7	7	2.5	7	6
Susceptibility tested to all 3 drugs	—	3	3	1.1	3	3
Resistance to 1 or more drugs—any degree	—	2	2	0.7	2	2
—definite	—	2	2	0.7	2	2
Remaining in hospital at 15 months after entry	6	29	35	12.7	13	7
Positive on culture at 15 months after entry	0	8	8	2.9	6	5
Susceptibility tested to all 3 drugs	—	4	4	1.4	3	3
Resistance to 1 or more drugs—any degree	—	3	3	1.1	3	3
—definite	—	3	3	1.1	3	3
Remaining in hospital at 18 months after entry	3	11	14	5.1	5	3
Positive on culture at 18 months after entry	0	4	4	1.4	3	2
Susceptibility tested to all 3 drugs	—	2	2	0.7	1	1
Resistance to 1 or more drugs—any degree	—	1	1	0.4	1	1
—definite	—	1	1	0.4	1	1

TABLE 9

ANALYSIS OF UNSATISFACTORY PROGRESS IN HOSPITAL

(First Admissions and Readmissions with Active Respiratory Tuberculosis Excluding Premature Discharges, Transfers out, and Nontuberculous Deaths)

	Total	Type of Admission		Extent of Pulmonary Tuberculosis			Cavity Definite on Entry	Bacteriologic Confirmation	Drug Susceptibility Tests on Entry						
										Less than Definite Drug Resistance only		Definite Drug Resistance			
		First Admissions	Readmissions	Minimal	Moderately Advanced	Far Advanced			Number Tested	1 Drug only	2 or More	1 Drug only (1—streptomycin) (1—isoniazid)	1 Drug and Less Resistance to Another	2 Drugs (streptomycin + isoniazid)	3 Drugs (streptomycin + PAS + isoniazid)
Totals	39	24	15	4	7	28	25	39	30	6‡	0	2	3	1	3
Deaths from tuberculosis	8*	4	4	0	0	8	5	8	5	0	0	0	0	0	1
Remaining in the hospital 18 months after entry	14	9	5	1	3	10	8	14	10	2	0	1	1	0	1
Result unsatisfactory at regular discharge	17	11	6	3	4	10	12	17	15	4	0	1	2	1	1
Open-positive cavity	1†	0	1	0	0	1	1	1	1	0	0	0	1	0	0
Closed-positive cavity	3†	2	1	0	2	1	2	3	3	0	0	0	0	1	1
Open-negative cavity	7	4	3	0	0	7	7	7	5	2	0	0	1	0	0
Roentgenogram continuing to clear only	6	5	1	3	2	1	2	6	6	2	0	1	0	0	0

* Average interval from entry to death = 30 days (shortest, 2 days; longest, 3 months).

† Three of these 4 patients had tubercle bacilli with definite drug resistance on entry and on discharge. All 4 patients were cooperative with maximal benefit from hospital care. Such discharges are made with prior approval of the local medical officer of health.

‡ 5 to streptomycin only; 1 to isoniazid only.

entry but were isolated three months later, drug-resistant strains were discovered in 4; but the discharge of drug-resistant bacilli was not continued. At six months after entry and thereafter, drug-resistant strains, if present among the patients remaining in the hospital, were usually definitely resistant. At six months after entry and thereafter, the patients who continued to discharge drug-resistant tubercle bacilli were usually those who had been discharging such bacilli at the time of hospital entry.

Analysis of unsatisfactory progress: Of 39 patients with unsatisfactory progress, 28 had far advanced pulmonary tuberculosis, 25 had definite cavitation on entry, and all 39 patients showed bacteriologic confirmation of their disease (table 9). On hospital entry, a total of 15 (50 per cent) of 30 patients in this group whose strains of bacilli were tested for drug susceptibility had strains with some degree of resistance to streptomycin, PAS, or isoniazid.

COMMENT

It is essential in reporting the incidence of strains of drug-resistant tubercle bacilli to define clearly the techniques and the criteria used. Standardization of bacteriologic technique and definitions for drug resistance would be most desirable.

The present report is concerned with the incidence of drug-resistant strains of tubercle bacilli observed in this group of patients who entered the hospital in 1957. Only through similar studies repeated in the future, with unchanging bacteriologic techniques and using the same definitions, will it be possible to know whether such drug resistance is increasing.

The unfavorable effect of drug-resistant tubercle bacilli upon therapeutic progress noted in this paper was apparent even with the use of concurrent triple-drug therapy in the vast majority of cases. It is reasonable to assume that on entry to the hospital, the effect upon

therapeutic progress of the presence of definite resistance to streptomycin, PAS, or isoniazid might have been more unfavorable had chemotherapy been limited to only two of the drugs mentioned. In other words, the use of concurrent triple-drug therapy makes it more likely that the organisms will be susceptible to two or at least one of the drugs used. Triple-drug therapy is therefore given some indirect support by these findings, particularly while awaiting the results of drug-susceptibility tests and particularly during that period of time for patients with pulmonary tuberculosis with cavitation, advanced disease, or a history of previous chemotherapy. Since early in 1957, it has been the custom in this hospital to omit streptomycin from the triple-drug regimen unless otherwise indicated at the time of regular discharge. Thus, most patients receive three drugs concurrently only while in the hospital, which now may represent only from one-third to one-half of the total duration of recommended therapy.

Summary

A study has been made of 497 patients consecutively admitted to the Toronto Hospital for Tuberculosis during the first eight months of 1957. A review of age, sex, familial descent, and country of birth confirmed that this group was reasonably representative of patients admitted to tuberculosis hospitals in recent years. An attempt was made to correlate various factors with therapeutic progress in the hospital during the eighteen months following entry for patients who were first admissions and readmissions with active pulmonary tuberculosis.

Among this group, the therapeutic progress was shown to be *least* satisfactory for those who entered as readmissions, had far advanced pulmonary tuberculosis, had definite cavitation, had tubercle bacilli in the sputum and drug-resistant tubercle bacilli, and were not suitable for pulmonary resection.

Perhaps the lesson is simply that there is no substitute in the management of tuberculosis for early diagnosis followed by prompt, carefully supervised, and adequate treatment accepted by a cooperative patient.

Sumario

Estudio de 500 Ingresos Consecutivos en un Hospital de Tuberculosis

Se ha ejecutado un estudio de 497 enfermos recibidos consecutivamente en el Hospital para Tuberculosis de Toronto durante los primeros ocho meses de 1957. Un repaso de la edad, sexo, descendencia familiar y país natal confirmó que este grupo era bastante típico de los enfermos admitidos en años recientes en los hospitales de tuberculosis. Tratóse de correlacionar varios factores con la evolución terapéutica en el hospital durante los dieciocho meses consecutivos al ingreso para los enfermos que eran primeros ingresos y reingresos con tuberculosis pulmonar activa.

En este grupo, se observó que la evolución terapéutica era lo *menos* satisfactoria para los admitidos como reingresos, que tenían tuberculosis pulmonar avanzada, tenían cavitación bien definida, ten an bacilos tuberculosos en el esputo y bacilos tuberculosos fármacorresistentes y no se prestaban para la resección pulmonar.

La lección aprendida quizás sea que, en la asistencia de la tuberculosis, no hay substituto para el diagnóstico temprano seguido del tratamiento pronto, vigilado cuidadosamente y adecuado, aceptado por un enfermo cooperativo.

Resume

Etude de 500 admissions consécutives dans un hopital de tuberculeux

L'auteur a pratiqué une étude de 497 malades admis à l'hopital des tuberculeux de Toronto, pendant les premiers huit mois de 1957. Considérant l'âge, le sexe, la descendance familiale et le pays de naissance ce groupe représente les malades admis dans les hopitaux pour tuberculeux ces dernières années. L'auteur a essayé de comparer des facteurs variant avec les progrés therapeutiques de l'hopital pendant les dix-huit mois qui ont suivi l'admission des malades dont c'était la première admission et les readmissions pour une tuberculose évolutive.

Parmi ce groupe, les progrés thérapeutiques ont été le moins satisfaisant pour ceux qui étaient des réadmissions, ceux qui étaient à un stade avancé de la maladie, ceux qui ayaient une caverne évidente, ceux qui avaient des bacilles dans leur expectoration, ceux qui avaient des bacilles résistant aux antibiotiques et ceux pour qui ne se posait pas l'indication de résection pulmonaire.

La leçon est peut-être simplement qu'il n'y a rien de plus efficace dans la façon de traiter une tuberculose, qu'un diagnostic précoce suivi rapidement d'un traitement correct, surveillé soigneusement et accepté par un malade qui veut coopérer.

Acknowledgment

The writer wishes to acknowledge the painstaking work of the physicians, nurses, technicians, stenographers, and other professional personnel in recording accurately the detailed clinical and laboratory information on the charts which he reviewed in this study.

CARDIOPULMONARY FUNCTION IN BRONCHIAL ASTHMA[1, 2]

A Comparison with Chronic Pulmonary Emphysema

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(Received for publication May 13, 1959)

INTRODUCTION

Previous studies have defined some of the abnormalities of pulmonary function that occur in bronchial asthma. Acute asthmatic attacks are characterized by severe airway obstruction, increased residual volume, and impaired mixing of gases in the lung, all of which abnormalities are reversible (1). During periods of remission, pulmonary function may be normal (2). Bates has pointed out that asthmatics have a normal uptake of carbon monoxide and that a relatively normal diffusing capacity distinguishes patients with asthma from those with emphysema (3). The present data were gathered in order to document further the type and degree of abnormality of cardiopulmonary function present in patients with bronchial asthma, and to ascertain whether or not pulmonary function in asthmatic patients with chronic dyspnea differs from that in patients with chronic obstructive emphysema.

MATERIALS AND METHODS

Fifteen patients with definite clinical diagnoses of bronchial asthma were studied. These patients reported attacks of wheezing and acute dyspnea, distinct from paroxysmal nocturnal dyspnea, precipitated by known allergens or respiratory infections and relieved by bronchodilators. Although the symptomatology of some patients at the time of the study consisted only of chronic, severe dyspnea, even these patients had typical histories of acute asthmatic attacks in the past, usually since childhood.

Ventilatory studies were performed before and after two to four inhalations of an aerosol bronchodilator.[4] Vital capacity and maximal midexpiratory flow rate (4) were calculated from the largest of at least two maximal expirations.

Arterial blood gas composition was measured by the Van Slyke manometric technique for oxygen and carbon dioxide contents; pH was measured on a Cambridge pH meter, and the carbon dioxide tension was calculated from a nomogram. Diffusing capacity for carbon monoxide was measured by the steady-state method (5).

Pulmonary vascular pressures and flows were measured by passing a polyethylene (6) or conventional nylon catheter into the pulmonary artery for blood sampling and recording of pressures. Values for systolic and diastolic pressures were obtained by averaging the measured pressures for one or two respiratory cycles and, similarly, mean pressures were electronically integrated over one to two respiratory cycles. Cardiac output was calculated from the Fick formula. Lung dead space (V_d) was calculated from the Bohr equation, using the arterial pressure of carbon dioxide as equivalent to the alveolar pressure of carbon dioxide, and the result was expressed as a fraction of the tidal volume (V_d/V_t).

Exercise studies were conducted in the supine position with the patient pedaling against a fixed resistance at the maximal rate that could be sustained for five minutes. Arterial blood and expired air were collected during the last two minutes of exercise.

RESULTS

The identifying data, clinical characteristics, and values for ventilatory function are shown in table 1. All of the patients revealed a variable degree of airway obstruction with, usually, a good response to bronchodilators. The vital capacity was moderately to severely reduced.

The data obtained during simultaneous study of arterial blood gas composition, diffusing capacity, and pulmonary circulation during rest and exercise are presented in table 2. The value for the maximal midexpiratory flow rate obtained on the day of study is also shown. There were, in general, mild hypoxemia and carbon dioxide retention which were only slightly increased during muscular exercise. The exercise diffusing capacity was subnormal in most of these patients but always above 11 cc. per min.

[1] From the Cardiorespiratory Laboratory, Grasslands Hospital, Valhalla, New York.

[2] Supported by grants from the Westchester Tuberculosis and Public Health Association, Westchester Heart Association, and the National Heart Institute, U. S. Public Health Service (Grant H-2437).

[3] Special Research Fellow, U. S. P. H. S., HF8347.

[4] Isoproterenol hydrochloride was supplied as Medihaler-iso through the courtesy of Riker Laboratories, Inc., Los Angeles, California.

TABLE 1

IDENTIFYING DATA, SYMPTOMS, AND VENTILATORY FUNCTION IN 15 CASES OF BRONCHIAL ASTHMA

(The mean value for maximal midexpiratory flow rate for each patient is listed along with the total range of values encountered, the number of measurements made, and the average per cent response (per cent improvement) after inhalation of a bronchodilator.)

Name	Age	Sex	BSA M²*	MMF L./Sec.†		N‡	Respiratory Bronchodilators	V.C.§	Clinical	
				Mean	Range				Duration	Severity
							per cent			
Ada	39	F	1.52	0.98	0.66–1.32	5	39	78	Childhood, 10 years	One flight SOBE‖
Ady	36	F	1.59	0.58	0.26–1.28	10	42	65	Life-long	Died in status
Car	56	M	2.14	0.75	0.49–1.04	5	33	64	2 years	Requires steroids
Dav	31	F	1.42	1.06	0.73–1.44	4	42	76	10 years	One to 2 attacks per month, relieved by bronchodilators
Doz	34	F	1.66	1.52	0.58–2.67	4	40	88	8 years	Nightly attacks
Har	36	F	1.66	1.48	0.20–3.80	9	41	85	2 years	One attack per month, relieved by bronchodilators
Kit	47	F	1.57	0.70	0.24–1.28	5	16	70	Life-long	Occassional attacks; sustained airway obstruction
McK	57	M	1.58	1.67	0.23–3.30	22	50	74	1 year	Severe attacks; requires steroids
Mor	65	M	1.82	0.52	0.31–0.76	21	25	65	Childhood, 10 years	Intermittent wheeze and SOBE
Qui	67	M	1.67	0.65	0.41–1.32	7	6	55	5 years	Sustained airway obstruction
Ruf	69	M	1.76	0.98	0.62–1.17	13	34	50	10 years	One flight SOBE
Sco	54	M	1.74	0.77	0.64–0.94	3	50	76	11 years	Slight SOBE
Ste	64	M	2.00	0.44		1	38	73	Life-long	SOBE; 1 flight limit
Tra	23	M	1.68	0.27	0.18–0.35	4	15	32	Life-long	Died in status
Wau	58	F	1.46	0.65	0.28–1.60	29	42	64	40 years	SOBE on one-half flight; occassionally regular steroids

* BSA M² = body surface area in square meters.
† MMF L./Sec. = maximal midexpiratory flow rate in liters per second.
‡ N = number of measurements.
§ V.C. = mean vital capacity.
‖ SOBE = short of breath on exertion.

per mm. of mercury. There was minimal pulmonary hypertension at rest, and a decided increase of the pulmonary arterial pressure occurred during exercise. The cardiac index was normal or elevated at rest and normal or reduced during exercise for the degree of oxygen consumption measured (7).

DISCUSSION

It is of interest to compare these findings in asthmatic patients with those in patients with chronic pulmonary emphysema (8). As a group (table 3), the patients with asthma revealed less impairment and greater variability of ventilatory function than the patients with emphysema. In addition, they showed greater improvement after the administration of a bronchodilator (9). The impairment of arterial blood gas composition was slightly greater in emphysema than in asthma, the degree of pulmonary hypertension during exercise was similar in the two groups, and the cardiac output was slightly higher in asthma than in emphysema. None of these differences was striking or distinctive, however, and a patient with asthma might exhibit the same abnormalities of ventilatory function, arterial blood gas composition, and pulmonary circulation as a patient with pulmonary emphysema. The one test which distinguished the two conditions was the diffusing capacity of the lungs. In patients with emphysema, the exercise diffusing capacity never exceeded 12.7 cc. per min. per mm. of

TABLE 2

VALUES FOR ARTERIAL CARBON DIOXIDE TENSION ($P_{a_{CO_2}}$), ARTERIAL OXYGEN SATURATION ($S_{a_{O_2}}$) OXYGEN CONSUMPTION ($\dot{V}_{O_2}$), CARBON MONOXIDE DIFFUSING CAPACITY (D_{CO}), PULMONARY ARTERIAL PRESSURE, CARDIAC INDEX, AND RATIO OF DEAD SPACE TO TIDAL VOLUME (V_d/V_t) DURING REST AND EXERCISE

(The maximal midexpiratory flow rate (MMF) recorded on the day of study is also listed.)

Name	State	$P_{a_{CO_2}}$	$S_{a_{O_2}}$	$\dot{V}_{O_2}$	D_{CO}	Pulmonary Arterial Pressure			Cardiac Index	V_d/V_t	MMF
						Systolic	Diastolic	Mean			
		mm. Hg.	*per cent*	*cc/min*	*cc/min. mm. Hg.*	*mm. Hg.*			*L./min./M²*	*per cent*	*L./sec.*
Ada	Rest	36	89	218	7.9					34	1.32
	Exercise	43	92	763	16.4					33	
Ady	Rest	48	90	258	9.6	43	31	38	3.26	51	0.30
Car	Rest	37	87	340	12.4					25	1.04
	Exercise	47	91	602	25.0					25	
Dav	Rest	29	92	169	11.4	21	7	12	3.30	34	1.30
	Exercise	29	91	575	15.6	32	12	20	5.08	23	
Doz	Rest	38	97	186	22.6					21	1.52
	Exercise	47	91	625	26.4					30	
Har	Rest	40	96	208	13.7					32	3.80
	Exercise	43	94	761	18.1					23	
	Rest	45	88	326	11.4					60	0.20
Kit	Rest	36	92	202	8.2	26	7	18	3.49	37	0.78
	Exercise	32	93	699	9.9	68	28	43	5.32	22	
	Exercise	45		798	14.3					35	
McK	Rest	41	91	291	13.2	15	6	12	5.31	39	3.94
	Exercise	46	91	915	19.4	49	35	43	6.65	36	
	Rest	52	80		11.9					50	0.25
Mor	Rest	44	90	275	12.8					39	0.42
	Exercise	47	89	629	14.4					37	
Qui	Rest	43	85	302	15.8	34	16	22	4.56	58	0.54
	Exercise	45	80	718	14.7					49	
	Rest	37	83	217	18.4	20	8	16	5.19	43	0.62
	Exercise	42	85	661	11.3	69	33	54	5.12	47	
Ruf	Rest	50	86	212	9.6	33			3.93	48	0.88
	Exercise	52	86	640	16.6	54			6.65	44	
Sco	Rest	53	93	259	26.3					41	0.64
	Exercise	50	93	688	39.6					34	
Ste	Rest	45	93	289	14.7					45	0.44
	Exercise	47	88	570	14.9					40	
Tra	Rest	70	84		18.0						0.18
Wau	Rest	37	92	239	11.2					39	0.61
	Exercise	38	93	417	14.2					29	
	Rest	39	92	223	13.7	32	8	22	3.56	38	0.52
	Exercise	38	87	463	12.0	43	14	21	4.62	33	

mercury, and was usually less than 10 cc. per min. per mm. of mercury (8). In the asthmatic patients reported above, the exercise diffusing capacity was rarely less than 13 cc. per min. per mm. of mercury, and was always above 11 cc. per min. per mm. of mercury.

As discussed elsewhere, the degree of reduction of the diffusing capacity in emphysema does not correlate with the degree of impairment of ventilatory function (8). The same is true in asthma, as shown by the data reported here. Furthermore, in 2 of the patients studied twice (*Har* and *McK*), the maximal midexpiratory flow rate changed markedly without significant change of the diffusing capacity, evidence that the level of the diffusing capacity is not greatly affected by the degree of ventilatory impairment. From these observations, it is reasonable to assume that the

TABLE 3

AVERAGE VALUES FOR SOME PARAMETERS OF CARDIOPULMONARY FUNCTION OBTAINED IN 15 CASES OF ASTHMA AND 22 PATIENTS WITH CHRONIC OBSTRUCTIVE PULMONARY EMPHYSEMA

(The total range of values encountered for the maximal midexpiratory flow rate is listed along with the average value for each group.)

	Asthma	Emphysema
Maximal midexpiratory flow rate (L./sec.)		
Mean	0.86	0.42
Range	0.20–3.80	0.16–1.00
Vital capacity (per cent)	<67	<54
Arterial oxygen saturation (per cent)		
Rest	89	86
Exercise	90	83
Arterial carbon dioxide tension (mm. Hg.)		
Rest	43	51
Exercise	43	53
Carbon monoxide diffusing capacity (cc./min./mm. Hg)		
Rest	13.9	7.1
Exercise	18.7	9.2
Cardiac index (L./min./M^2)		
Rest	3.96	3.32
Exercise	5.61	4.22
Pulmonary arterial pressure (mm. Hg)		
Rest	28/12	35/14
Exercise	52/24	51/22

diffusing capacity of the lung is a measure of the alveolar capillary surface area and is largely independent of the state of the lung airways (unless complete airway obstruction leads to collapse of alveoli). Thus, in either asthma or emphysema, the extent of destruction of alveoli may be reflected in the degree of reduction of the diffusing capacity. When destruction of alveoli is minimal, as in asthma, there is slight or no reduction of diffusing capacity for carbon monoxide. When destruction of alveoli is marked (the major pathologic criterion of emphysema), there is severe impairment of diffusing capacity for carbon monoxide. Establishment of the validity of this concept requires correlation between pathologic studies and measurements of the diffusing capacity.

Whether or not bronchial asthma eventually leads to sufficient destruction of alveolar capillaries to meet the pathologic criteria of emphysema remains to be decided. Some of the asthmatic patients reported above had long-standing airway obstruction yet relatively normal diffusing capacities, suggesting absence of emphysema. However, the low diffusing capacity measured in others suggests the presence of some emphysema.

The pulmonary hypertension found in many of these patients when they were free of acute asthmatic symptoms is of some interest and suggests that there is increased resistance to blood flow through the lungs. Previous studies have revealed a decrease of this resistance to blood flow after the administration of atropine. Atropine had no such effect on the pulmonary circulation of most of the patients with emphysema who were studied (10). These studies suggested that pulmonary hypertension in asthma is largely the result of increased intra-alveolar pressure secondary to airway obstruction. In contrast, the pulmonary hypertension in emphysema is thought to result from actual loss of pulmonary vessels, with consequent increase of pulmonary vascular resistance. In both diseases severe hypoxemia may aggravate pulmonary hypertension and lead to the development of cor pulmonale (11).

The patient with bronchial asthma may suffer the same disabling shortness of breath as the patient with emphysema. In both diseases, dyspnea is due to the increased work required to move air through the obstructed airways. Despite the fact that airway obstruction may become severe enough to cause death in status asthmaticus, the prognosis in asthma is relatively good since the obstruction is usually reversible. In contrast, the prognosis in emphysema is considerably less favorable. The airway obstruction is largely irreversible, and the loss of alveolar capillaries causes fixed pulmonary hypertension during exercise which predisposes to the development of cor pulmonale.

SUMMARY

Measurements were made of ventilatory function, arterial blood gas composition, diffusing capacity, and pulmonary circulation in 15 patients with bronchial asthma. There was a variable degree of airway obstruction, hypoxemia, carbon dioxide retention, pulmonary hypertension, and slight reduction of the diffusing

capacity. The results are contrasted with those observed previously in chronic pulmonary emphysema. The only test which consistently distinguished between the two diseases was the diffusing capacity. This was always severely reduced in emphysema, but normal or only moderately reduced in asthma. This finding is interpreted as a reflection of destruction of alveolar capillaries in emphysema and a relatively normal alveolar capillary surface in asthma.

Sumario

La Función Cardiopulmonar en el Asma Bronquial: Comparación con el Enfisema Pulmonar Crónico

En 15 enfermos de asma bronquial, se hicieron mediciones de la función ventiladora, la composición del gas de la sangre arterial, la capacidad difusora y la circulación pulmonar. Se observó en grado variable oclusión de las vías aéreas, hipoxemia, retención del bióxido de carbono e hipertensión pulmonar y leve disminución de la capacidad difusora. Se contrastan los resultados con los observados antes en el enfisema pulmonar crónico. La única prueba que diferenció constantemente las dos enfermedades fué la capacidad difusora. Esta estuvo siempre intensamente reducida en el enfisema, pero fué normal o no estuvo más que moderadamente reducida en el asma. Se interpreta esta hallazgo como reflejo de la destrucción de los capilares alveolares en el enfisema y de una superficie relativamente normal de los capilares alveolares en el asma.

Resume

La fonction cardiopulmonaire dans l'asthme bronchique. Comparaison avec l'emphysème pulmonaire chronique

On a mesuré la fonction de ventilation, la composition gazeuse du sang artériel, la capacité de diffusion et la circulation pulmonaire chez quinze malades ayant de l'asthme bronchique. On trouva un degré variable d'obstruction des voies respiratoires, de l'hypoxie, de la rétention de gaz carbonique, de l'hypertension pulmonaire et une légère diminution de la capacité de diffusion. Les résultats s'opposent à ceux observés jusqu'à présent dans l'emphysème pulmonaire chronique. Le seul résultat qui différencia d'une façon sure les deux maladies fut la capacité de diffusion. Cette dernière fut toujours considérablement diminuée dans l'emphysème, mais normal ou seulement modérèment diminuée dans l'asthme. Cette découverte est interprètée comme le résultat de la destruction des capillaires alvéolaires dans l'emphysème, alors que, dans l'asthme, la surface des capillaires alvéolaires est relativement normale.

Acknowledgment

The careful technical assistance of Cecile Kane and Catherine Doyle is gratefully acknowledged.

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ELECTROCARDIOGRAPHIC EFFECTS OF PNEUMOPERITONEUM[1]

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(Received for publication June 16, 1959)

INTRODUCTION

Artificial pneumoperitoneum may produce electrocardiographic changes of sufficient degree to arouse serious suspicion of myocardial injury. Prior investigations have been concerned largely with electrocardiographic changes found shortly after the injection of air into the peritoneum, or with the reversal of changes immediately following withdrawal of air. Studies of the electrocardiographic changes resulting from artificial pneumoperitoneum alone (1–7) or combined with other procedures, e.g., phrenic crush (8–10), capable of altering the electrocardiogram independently, have been based upon inspection of the tracings. There have been no studies of ventricular gradients ($\hat{G}$) from electrocardiograms influenced by artificial pneumoperitoneum alone, although data on the manifest axis of QRS ($\hat{A}QRS$) were given from twenty-five such cases (6) and for $\hat{A}QRS$ and $\hat{G}$ from five cases with combined artificial pneumoperitoneum and unilateral phrenic crush (10).

The purpose of the present study is to present the electrocardiographic data on patients maintained on pneumoperitoneum alone over periods of many weeks. The data indicate the incidence of change in the tracings, the nature of the changes encountered, and analysis of $\hat{A}QRS$ and $\hat{G}$ in all instances in which any electrocardiographic change or abnormality was noted either before or after artificial pneumoperitoneum.

MATERIAL AND METHODS

Thirty-two patients suffering from pulmonary tuberculosis were given artificial pneumoperitoneum after an initial control standard twelve lead electrocardiogram had been obtained. All tracings were taken in the recumbent position in view of the influence of the upright position noted by others (6, 8). All of the patients had far advanced pulmonary tuberculosis. The amount of air injected into the peritoneal cavity and the adequacy of the pneumoperitoneum to effect and maintain elevation of the diaphragms was controlled by roentgenographic inspections. Attempts to assess changes in cardiac position from roentgenographic data were considered too unreliable to include in the study. Changes in the status of pulmonary disease during the period of artificial pneumoperitoneum were not deemed capable of altering the electrocardiogram.

After artificial pneumoperitoneum had been well established, another electrocardiogram was obtained. The usual interval was six to eight weeks, in a few from three to six weeks, with a maximum of twelve weeks. In 12 instances in which the electrocardiogram varied in any way from a normal pattern either before or after artificial pneumoperitoneum, $\hat{A}QRS$ and $\hat{G}$ were derived according to standard methods (11–17).

OBSERVATIONS

Analysis of Electrocardiograms

Of the 32 patients in this study, 20 showed no abnormalities of the electrocardiogram before or after artificial pneumoperitoneum. The only differences noted in these were shifts in the axes in a few instances. In the other 12 cases, sufficient changes were displayed to warrant detailed analysis which included determinations of the manifest axis of QRS and the ventricular gradient. While deviations from normal were noted in various leads in some of these cases, lead III displayed some change in all 12 instances. Therefore, all significant electrocardiographic deviations of QRS and T seen in lead III either before or after artificial pneumoperitoneum were recorded (table 1). When no significant deviations from normal standards were seen, no record appears in the table. Marked changes in amplitude of the QRS complex were seen occasionally, but these were not shown if due to axis shift alone. The cardiac axes derived from inspection of lead aVl and aVf and the transition zone of the precordial QRS were noted. The pre- and post-artificial pneumoperitoneum angles and magnitudes of $\hat{A}QRS$ and $\hat{G}$, respectively, as well as the ventricular rates of these twelve tracings, appear in the table.

In *lead III*, a Q wave appeared after artificial pneumoperitoneum in 4 patients, and a preexisting Q-wave became more pronounced in another.

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TABLE 1

SIGNIFICANT ELECTROCARDIOGRAPHIC DEVIATIONS BEFORE AND AFTER PNEUMOPERITONEUM

Case	Lead III				1		2		3				4				5	
	QRS		T		Axis		Tr. Zn.		ÂQRS				Ĝ				Rate	
	B	A	B	A	B	A	B	A	B		A		B		A		B	A
1		Q$\mathcal{S}$	↓	↓	SV	V	5	6	0°	2.2	59°	5.5	8°	6.4	25°	8.4	65	69
2		q$\mathcal{S}$	Iso	↑	INT	INT	4	3	53°	4.0	48°	4.5	45°	7.5	45°	9.8	105	75
3			↓	↓	SV	SV	2	2	70°	6.2	56°	5.0	41°	9.0	22°	8.4	86	90
4	M	rsR′	↑	Iso	SV	SH	3	4	62°	3.0	44°	4.8	50°	7.0	24°	7.5	86	88
5	q	Q	~	↓	SV	SV	2	1	50°	3.6	33°	6.0	29°	9.0	24°	12.0	112	88
6	Rs	rsR′	↑	↓	INT	SV	4	2	40°	3.6	34°	5.0	43°	9.5	31°	14.7	82	60
7			↓	↑	V	V	4	4	72°	13.4	62°	14.0	67°	12.8	53°	13.0	62	88
8		q$\mathcal{S}$			SV	V	4	5	−14°	2.2	68°	6.0	36°	5.5	60°	15.6	96	60
9			↑	↑	SV	INT	3	2	56°	17.5	38°	10.5	53°	14.0	42°	11.8	115	88
10			Tall	Tall	V	V	4	4	80°	6.4	72°	6.6	78°	18.5	69°	11.5	98	105
11			↓	↓	V	V	4	4	54°	10.4	53°	11.2	36°	15.2	34°	15.0	105	90
12		Q	↓	↓	V	V	6	6	74°	5.5	67°	5.0	53°	12.6	56°	12.5	88	92

The left-hand columns show the significant electrocardiographic findings for QRS and T noted on inspection of *LEAD III ONLY*. Changes in magnitude of QRS, if otherwise not remarkable, are not indicated. Sub-columns B and A refer to data found *before* and *after* artificial pneumoperitoneum, respectively. Column 1 shows estimated frontal plane axis based on inspection of aVl and aVf. In column 2, the number indicates the transition zone of precordial QRS. Column 3 shows the direction of ÂQRS in degrees on the triaxial reference scale and its length in units (1 unit = 4 microvolt seconds), respectively, in paired columns. Column 4 gives similar data for Ĝ; and column 5, ventricular rates before and after artificial pneumoperitoneum.

q = small Q wave
$\mathcal{S}$ = disappearance of S wave
Arrows indicate relative size and direction of T-waves
V = vertical
H = horizontal
INT = intermediate
ÂQRS = manifest axis of QRS
Ĝ = ventricular gradient
Q = large Q wave
SV = semivertical
SH = semihorizontal

A Q_3T_3 pattern developed in 3 instances. In 2 other cases there were changes in R and S without the development of Q-waves. The S-T segment was initially elevated in 2 patients and depressed in 2, and became isoelectric in one instance in each category. In 7 cases T_3 was initially inverted; it became deeper in 3 of these, became upright in one, and remained the same in 3. When T_3 was initially upright or isoelectric, it became taller in one case, inverted in one case, and lower in 2 cases, remaining unchanged in the rest.

In *lead aVf*, changes in QRS paralleled those in lead III, but were less pronounced. T-wave changes occurred as follows: in *Case 1*, the T-wave went from isoelectric to low upright, the opposite of the findings in lead III; in *Case 3*, the T-wave went from tall upright to low upright while, in lead III, T changed from small inverted to deeply inverted; in *Case 7*, the T-wave went from small inverted to deeply inverted where lead III went from inverted to upright. The TaVf in *Cases 9*, *10*, and *11* were very similar to those seen in lead III.

Lead II failed to reveal S-T segment shifts as a result of artificial pneumoperitoneum, as noted by other writers. It was depressed in *Case 9* and elevated in *Cases 10* and *12*, both before and after artificial pneumoperitoneum. An upright T_2 became taller in *Case 1*, and in *Case 7* preartificial pneumoperitoneum T inversion remained unchanged.

In *lead aVl* an upright T became taller in *Case 7*, and an isoelectric T-wave became upright in *Case 8*. In *Cases 9* and *10*, T was found similarly inverted before and after artificial pneumoperitoneum.

Leads I and *aVr* were affected only in *Cases 7* and *9* and will be discussed below.

Precordial lead changes after artificial pneumoperitoneum showed increased negativity of T in

lead V_2 in 4 cases. The transition zone of QRS in the precordial leads was found in V_2 in 3 cases, and in V_6 in 3 others. Artificial pneumoperitoneum did not alter these significantly. Shifts in the transition zone between leads V_4, V_5, and V_6 were noted in some of the other cases. In addition, in *Case 10*, all precordial T-waves were tall and peaked as they appeared in leads II, III, and aVf. In *Case 12*, terminal inversion of T appears in V_2-V_6 after artificial pneumoperitoneum. This electrocardiogram showed marked clockwise rotation in the vertical axis. Precordial lead changes in *Cases 7* and *9* will be discussed below.

The *axis of the heart* as gauged by inspection of leads aVl and aVf before and after artificial pneumoperitoneum is shown in the first column of table 1. In 7 of the 32 cases this shifted toward a more vertical orientation, in 5 cases it shifted toward the horizontal, and it remained unchanged in 20. In *Cases 1*, *6*, and *8* the findings in column 1 are not congruent with those in columns 3 and 4, showing $\hat{A}QRS$ and $\hat{G}$. In these cases, displacement of the heart and rotation about other axes account for this inconsistency.

The *QRS vector* had normal direction in all instances. After artificial pneumoperitoneum it was seen to rotate through an angle of 6° to 18° to the left in 9 instances, and to the right 59°, 82°, and 1°, respectively, in the remaining cases. The magnitude of $\hat{A}QRS$ lay within normal ranges both before and after artificial pneumoperitoneum and the changes were consistent with rotational changes noted.

The *ventricular gradient* rotated through an angle varying from 2° to 26° leftward in 8 instances; remained unchanged in one; and rotated rightward 17°, 24°, and 3°, respectively, in the remaining instances. Its direction was within normal bounds, and magnitudes were in the normal limits of plus 23 to plus 2.5 units. Normally, $\hat{G}$ lies not more than 35° to the left nor more than 24° to the right of $\hat{A}QRS$. In *Case 9*, before artificial pneumoperitoneum, $\hat{G}$ was 50° to the right, but afterward it was found 8° to the left of $\hat{A}QRS$. In all other instances, the relationship of the angle of $\hat{A}QRS$ and $\hat{G}$ was normal.

Changes in the *P-wave* were noted in 2 cases. In *Case 2* it was tall and peaked in leads II and aVf initially, and became lower after artificial pneumoperitoneum. In *Case 11*, after artificial pneumoperitoneum only, in the limb leads inverted and upright-P waves appeared in random fashion. Inversion of the P-wave was associated with shortening of P-R to 0.12 seconds or less, depression or "coving" of P-R, and prolongation of QRS to 0.10 seconds. Where P was upright, P-R was isoelectric and 0.14 to 0.16 seconds, and QRS was 0.6 seconds duration. All precordial P-waves were inverted.

Illustrative Cases

Case 7: A middle-aged diabetic presented the following electrocardiographic findings before artificial pneumoperitoneum: cardiac rate, 62 per minute; normal sinus rhythm; P-R=0.20; QRS=0.06; Q-T=0.30 seconds. T in leads I, II, aVr, and aV1, low and upright; in III and aVf, inverted: QRS in V_4 and V_5, splintered. After artificial pneumoperitoneum: cardiac rate, 88 per minute without change in intervals; T_1 unchanged; T_3 became upright while TaVf became deeper; and T in aVr, aV1, and II became taller. $T_{V_2\text{-}V_6}$ became inverted with slight elevation and coving of S-T. Because hyperglycemic influences were suspected, another electrocardiogram was done one month later when the diabetes was well controlled and the artificial pneumoperitoneum maintained. Precordial T-wave inversion persisted but was less marked, and S-T segment elevation was still noted. All limb lead T-waves were normally directed at rest, but were reversed during exercise. In spite of these changes, ventricular gradients were well within normal limits before and after artificial pneumoperitoneum.

Case 9: A middle-aged female without evidence of cardiac disease presented electrocardiographic findings before artificial pneumoperitoneum as follows: cardiac rate, 115; normal sinus mechanism; P-R=0.16; QRS=0.08; and Q-T=0.32 seconds. T_1, aVr, aV1 were small and terminally negative. $S\text{-}T_{II}$ and $S\text{-}T_{III}$ were slightly depressed with small, upright T, and small upright T_{aVf}. T in $V_{1\text{-}5\text{-}6}$ was negative, and in $V_{2\text{-}3\text{-}4}$ biphasic, with coving of S-T throughout. After artificial pneumoperitoneum: cardiac rate, 88 with P-R and QRS intervals unchanged, and Q-T=0.36 seconds.

Aside from slight counterclockwise rotation, in horizontal and vertical axes, the changes noted before persisted but diminished, shifting toward the normal. Ventricular gradients were within normal limits both before and after artificial pneumoperitoneum.

Discussion

Pneumoperitoneum may produce changes in the electrocardiogram suggesting the possibility of cardiac disease, although clinical evidence is lacking. Since artificial pneumoperitoneum produces no clinical evidence of cardiac disorder, all writers have postulated electrocardiographic changes on the basis of changed heart position.

Interpretation of changes in cardiac position by inspection of QRS in the limb leads indicates considerable variability from artificial pneumoperitoneum. In the present study, artificial pneumoperitoneum produced shift to a more vertical axis in 7 cases, and toward a more horizontal axis in 5, the remaining 20 cases being unchanged. Other writers report a somewhat greater percentage of cases showing axis shift and a larger proportion toward the horizontal.

In an effort to clarify further the importance of electrocardiographic changes in artificial pneumoperitoneum, an analysis was undertaken of the manifest QRS vector and the ventricular gradient in the 12 cases showing electrocardiographic changes before or after artificial pneumoperitoneum. There was a leftward shift of $\hat{A}QRS$ in 9 of the 12 cases (although of minute degree in 3) in the present study, from an average of plus 62° before to plus 50° after artificial pneumoperitoneum, and 2 patients showed marked shifts to the right. There was no change in one. Carasso and co-workers (6) plotted $\hat{A}QRS$ in twenty-five cases and found a leftward shift in all from an average of plus 56.8° before to plus 31.8° after artificial pneumoperitoneum.

Of greater significance is the ventricular gradient. Substantial change in the angle formed by $\hat{A}QRS$ with $\hat{G}$ is indicative of rotation about the vertical axis and was noted in *Cases 1* and *8*. In the latter, the added influence of difference in rate (16) upon the length of $\hat{G}$ is seen here as well as in *Cases 2*, *5*, and *6*. In *Case 9*, $\hat{G}$ becomes unaccountably shortened in spite of slower rate and insignificant rotation about the vertical axis, and in *Case 10*, $\hat{G}$ is shortened with no change of rate or vertical axis. In all of the cases, the magnitude of $\hat{G}$ remained within normal limits.

While artificial pneumoperitoneum will produce leftward shift of $\hat{A}QRS$ in 75 per cent of cases, there is usually little effect upon $\hat{G}$. A change in the magnitude of $\hat{G}$ without change of direction may be seen as a result of rotation about the vertical axis, change in rate, or nonpathologic influences, as pointed out by Rosen and Gardberg (18). On the basis of the present study, artificial pneumoperitoneum would qualify for inclusion in this group of nonpathologic causes for electrocardiographic changes.

Benatt and Berg (10) derived ventricular gradients in five cases of their series, but their findings are not strictly comparable with those of the present study since phrenic crush was combined with artificial pneumoperitoneum. Nevertheless, their data indicate rotation of the heart mainly in an anticlockwise direction about the horizontal axis in the saggital plane, i.e., elevation of the apex and depression of the base. They conclude that artificial pneumoperitoneum entails no danger to the cardiovascular status of patients with normal hearts.

Reports on the effect of artificial pneumoperitoneum alone upon the electrocardiogram have emphasized most commonly Q- and T-wave changes in lead III. The reported incidence of these, as well as other less frequently seen changes, has varied widely. In the present study, a Q-wave in aVf was present whenever Q_3 was seen. In no case was Q in aVf of greater magnitude than Q_3 either before or after artificial pneumoperitoneum.

Aside from the fairly extensive T-wave changes noted in the 2 cases which were presented in detail, the bulk of T-wave changes was confined to lead III. In the 3 instances of alterations of T-wave in lead aVf, they were opposite to those noted in lead III. Under all circumstances, the true significance of the deviations of the T-wave became apparent when the ventricular gradient was derived and found to be within normal limits.

Shift of the S-T segment, especially in lead II, has been reported. The present study and that of Sreenivasan (19) particularly note no significant changes.

Since the electrocardiograms in the present study were done after a relatively long period of established artificial pneumoperitoneum, it is possible that there was an "accommodation" by the thoracic structures so that the heart is less likely to be found displaced the longer artificial pneumoperitoneum is maintained. Indeed, this accommodation may be the reason for the disappointing results of artificial pneumoperitoneum therapy and its recent loss of status in the treatment of pulmonary tuberculosis.

Summary

Standard twelve-lead electrocardiograms taken with the subject recumbent were made before and after induction of pneumoperitoneum in 32 patients with pulmonary tuberculosis. Pneumoperitoneum was established from three to twelve weeks before the second tracing was made.

Changes in Q_3 appeared in 5 cases accompanied by similar but lesser changes in lead aVf. Inversion of T_3 occurred in 3 of these cases, but

T changes in aVf appeared unrelated. Except for amplitude differences, QRS complexes and T-waves were normal in the limb leads in the other cases, except for 2 cases discussed in detail.

Shifting of the transition zone and T-wave changes in the precordial leads were accounted for by rotation of the heart about the vertical axis and/or mediastinal shift.

In 2 cases showing abnormal electrocardiographic patterns before pneumoperitoneum, detailed analysis indicated no adverse effect by electrocardiographic or clinical criteria.

The manifest axis of QRS and the ventricular gradient were determined in the 12 patients of the series who showed any electrocardiographic change. While estimates from inspection of leads aVl and aVf indicated a shift toward a more vertical axis in 7 patients and toward the horizontal axis in 5 of the 32 cases, the ventricular gradient deviated leftward in 8 instances, rightward in 3 cases, and remained unchanged in one.

In the presence of electrocardiographic changes following pneumoperitoneum, ventricular gradients remain within normal limits and preserve normal relationships with ÂQRS. This indicates that pneumoperitoneum causes change in heart position or diminution in magnitude of the ventricular gradient, such as is seen in a variety of nonpathologic conditions.

Sumario

Efectos Electrocardiográficos del Neumoperitoneo

En 32 tuberculosos pulmonares, se obtuvieron, con el enfermo recostado, electrocardiogramas corrientes de doce derivaciones antes y después de la ejecución del neumoperitoneo. Este se hallaba establecido de tres a doce semanas antes de obtener el segundo trazado.

En 5 casos, aparecieron alteraciones en la Q_3, acompañadas de alteraciones semejantes, pero menores, en la derivación aVf. En 3 de estos casos, hubo inversión de la T, pero las alteraciones de la T en la aVf no guardaban al parecer relación. Exceptuando las diferencias en la amplitud, los complejos QRS y las ondas T fueron normales en las derivaciones de los miembros en los demás casos, aparte de 2 casos discutidos a fondo.

La desviación de la zona de transición y las alteraciones de la onda T en las derivaciones precordiales quedaron explicadas por la rotación del corazón alrededor del eje vertical y/o la desviación del mediastino.

En 2 casos que revelaron patrones electrocardiográficos anómalos antes del neumoperitoneo, un análisis pormenorizado no indicó efecto adverso a base de pautas electrocardiográficas o clínicas.

En los 12 enfermos de la serie que revelaban alguna alteración electrocardiográfica, se determinaron el eje manifiesto de la QRS y la pendiente ventricular. Aunque los cálculos basados en la inspección de las derivaciones aVl y aVf indicaban desviación hacia un eje más vertical en 7 enfermos y hacia el eje horizontal en 5 de los 32 casos, la pendiente ventricular se desviaba hacia la izquierda en 8 casos, hacia la derecha en 3 y permaneció inalterada en uno.

Cuando se observan alteraciones electrocardiográficas consecutivamente al neumoperitoneo, las pendientes ventriculares permanecen dentro de límites normales y retienen sus relaciones normales con la ÂQRS. Esto indica que el neumoperitoneo ocasiona alteraciones en la posición del corazón o disminución en la magnitud de la pendiente ventricular, tal como se observa en varios estados que no son patológicos.

Resume

Rétentissement electrocardiographique du pneumopéritoine

Des électrocardiogrammes standard, sur sujet étendu, ont été pratiqués avant et aprés création d'un pneumopéritoine chez 32 malades ayant une tuberculose pulmonaire. Le pneumoperitoine fut établi de 3 a 12 semaines avant un second tracé.

Il apparut des changements de Q_3 dans 5 cas, accompagnés de changements moins importants de aVf. L' inversion de T_3 s'est produit dans 3 des cas, mais les modifications de T dans aVf ne semblèrent pas être en relation. A part une différence d' amplitude, les complexes QRS et les ondes T furent normaux dans les dérivations des membres chez d'autre cas, excepté 2 cas qui sont décrits en détail.

Le déplacement de la zone de transition et des changements de l'onde T dans les dérivations précordiales sont attribués à la rotation du coeur autour de l'axe vertical ou à un déplacement mediastinal.

Dans 2 cas ayant des tracés anormaux avant le pneumopéritoine une analyse méticuleuse ne montra aucun effet contraire, d'après les critères electrocardiographiques et cliniques.

Le complexe QRS et les tracés ventriculaires furent étudiés chez 12 des malades qui montrèrent un changement electrocardiographique. L'étude des dérivations aVl et AVf indiqua un déplacement vers un axe plus vertical chez 7 malades et vers l'axe horizontal dans 5 des 32 cas. Le complexe ventriculaire fut devié vers la gauche dans 8 cas, vers la droite dans 3 cas et resta inchangé dans un cas.

Quand il existe des changements electrocardiographiques aprés un pneumoperitoine, les complexes ventriculaires restent dans les limites de la normale et conservent des proportions normales par rapport à ÂQRS. Ceci indique que le pneumoperitoine produit un changement de la position du coeur ou une diminution d'amplitude du trace ventriculaire, comme cela se voit dans certaines conditions nonpathologiques.

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PNEUMONECTOMY IN THE TREATMENT OF PULMONARY TUBERCULOSIS[1, 2, 3]

The After History

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(Received for publication May 19, 1959)

INTRODUCTION

Important to clinicians considering pneumonectomy in the treatment of tuberculosis are such ponderables as the percentage of patients who will be dead, will have active disease, or will have inactive disease at a stated number of years after the operation. The present study examines the outcome of pneumonectomy in 204 patients undergoing treatment for pulmonary tuberculosis at Firland Sanatorium, Seattle, during a nine-and-a-half-year period. The patients came under study at the time of their pneumonectomies and were placed in three groups: (*a*) those who had had previous thoracoplasties, (*b*) those who had had concomitant thoracoplasties, and (*c*) those who had not had thoracoplasties. The three groups were analyzed separately and collectively by the anniversary method (1).

METHODS

The selection of cases for surgery and the details of the sanatorium and post-sanatorium course were previously described (2). The period of the study, January 1, 1946, to June 30, 1955, includes the "adequate chemotherapy era" (1953–1955). Sixteen pneumonectomies without previous or concomitant thoracoplasties were completed during this era. Application of the anniversary method to this group separately reveals no significant deviation from the others for the first two years of the post-pneumonectomy state. Therefore, these cases are included in the total number without distinction. For the most part, the remainder of the patients, whose pneumonectomies had been performed before 1953, had not received chemotherapy for more than six weeks.

At the time of surgery and at yearly intervals (anniversaries) thereafter, each patient was assigned to one of the following categories.

Inactive disease: For six months or more prior to the anniversary the patient's sputum had been free of tubercle bacilli, and his serial chest roentgenograms had been compatible with a diagnosis of stable disease.

Active disease: Within six months prior to the anniversary the patient's sputum contained tubercle bacilli, or serial chest roentgenograms had shown unstable disease.

Died of tuberculosis: During the year prior to the anniversary the patient died of tuberculosis. This category includes deaths from progressive tuberculosis and from causes related to the pneumonectomy.

Died of nontuberculous causes: During the year prior to the anniversary the patient died of causes known to be other than tuberculosis or sequelae of the operation.

Lost: The patient's whereabouts became unknown during the year preceding the anniversary.

Incomplete: The patient was alive and traced at the previous anniversary but had too short a follow-up study to attain the specified anniversary.

A patient might be assigned to the active or inactive category a number of times, depending upon the postoperative course of his disease and his availability for further study, as well as on the amount of time elapsing between his operation and the end of the year. Assignment to any of the last four categories, of course, terminated the patient's inclusion in the study.

RESULTS

Use of the annversary method (table 1) shows that pneumonectomy in the treatment of 204 cases of tuberculosis resulted in 60 per cent of the patients having inactive disease one year after surgery. By the second anniversary, almost 70 per cent had inactive disease. After the third anniversary, less than 2 per cent had active disease. By the first anniversary, 10 per cent were dead of tuberculosis or from causes related to the pneumonectomy, and 21 per cent were dead on the eighth anniversary of their surgery.

[1] From Firland Sanatorium, and the Department of Medicine, University of Washington, School of Medicine, Seattle, Washington.

[2] This study was aided by a grant from the Committee on Medical Research of the American Trudeau Society, medical section of the National Tuberculosis Association.

[3] Presented in part before the Medical Session, as part of *Section 3A*, at the annual meeting of the American Trudeau Society, Kansas City, Missouri, May 7, 1957.

TABLE 1

PATIENTS TREATED BY PNEUMONECTOMY AT THE FIRLAND SANATORIUM, 1946–1955

Classified by Status at Each Anniversary

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)	(11)	(12)	(13)	(14)	(15)
Anniversary	Living at Anniversary		Since Previous Anniversary			Total	Incomplete	Factors		Expected Percentages				
	Inactive	Active	Died of Tuberculosis	Died of Nontuberculous Disease	Lost			Provisional	Correction	Inactive	Active	Died of Tuberculosis	Died of Nontuberculous Disease	Lost
0	—	204	—	—	—	204	—	1.0000	0.4902	—	100.0	—	—	—
1	118	47	21	5	6	197	7	1.0355	0.5076	59.90	23.86	10.66	2.54	3.06
												10.66	*2.54*	*3.06*
2	134	15	7	1	6	163	3	1.0184	0.5169	69.26	7.75	3.62	0.52	3.10
												14.28	*3.06*	*6.16*
3	124	10	2	2	2	140	8	1.0571	0.5464	67.75	5.46	1.09	1.09	1.09
												15.37	*4.15*	*7.25*
4	107	3	2	2	2	116	18	1.1552	0.6312	67.54	1.89	1.26	1.26	1.26
												16.63	*5.41*	*8.51*
5	82	2	1	—	2	87	23	1.2644	0.7980	65.44	1.60	0.80	—	1.60
												17.43	*5.41*	*10.11*
6	40	1	—	—	—	41	43	2.0489	1.6350	65.40	1.64	—	—	—
												17.43	*5.41*	*10.11*
7	17	—	1	—	—	18	23	2.2778	3.7242	63.31	—	3.72	—	—
												21.15	*5.41*	*10.11*
8	4	—	—	—	—	4	13	4.2500	15.8278	63.31	—	—	—	—
												21.15	*5.41*	*10.11*

(1) Number of years after pneumonectomy.
(2) Number of living patients with inactive disease at anniversary.
(3) Number of living patients with active disease at anniversary.
(4) Number who died of tuberculosis since previous anniversary.
(5) Number who died of causes other than tuberculosis since previous anniversary.
(6) Number lost since previous anniversary.
(7) Total number at anniversary, sum of columns (2) to (6) inclusive.
(8) Number of "incomplete" patients at anniversary. These patients were alive and traced at the previous anniversary, but had too short a follow-up study to attain the specified anniversary.
(9) Provisional factor for anniversary— $\frac{\text{column (7) plus column (8)}}{\text{column (7)}}$
(10) Correction factor for anniversary—column (9) times correction factor for previous anniversary. The correction factor for anniversary 0 is 100/204 or 0.4902.
(11) Expected percentage of living patients with inactive disease at anniversary: column (2) times column (10).
(12) Expected percentage of patients with active disease at anniversary: column (3) times column (10).
(13) Expected percentage of patients dead of tuberculosis at anniversary: column (4) times column (10). Cumulative percentages in italics.
(14) Expected percentage of patients dead of causes other than tuberculosis at anniversary: column (5) times column (10). Cumulative percentages in italics.
(15) Expected percentage of patients lost at anniversary: column (6) times column (10). Cumulative percentages in italics.

One hundred and thirty-four patients (66 per cent) in this study had pneumonectomies with thoracoplasties following the resection within two weeks. Included in this grouping are patients who were chosen to have thoracoplasties following their pneumonectomies, but who died in surgery or postoperatively before thoracoplasty could be completed. That the results

might not be biased by the omission of these cases, they are included in the pneumonectomy with concomitant thoracoplasty group. When the entire group was compared with the others, there was no significant difference at any anniversary in the number expected to have active disease or inactive disease, or to have died of tuberculosis.

Fifty-four (26 per cent) of the pneumonectomies were completed after the failure of a thoracoplasty to control the disease. These patients, also, were not significantly different from the remainder in any category, although the percentage of those with inactive disease and those dead of tuberculosis ran somewhat higher at each anniversary than is shown in table 1.

Sixteen of the patients (8 per cent) had no thoracoplasty either before or after the pneumonectomy. The operations on these patients were performed after 1952, and only 3 were available for the third anniversary. Little difference in the status of the 16 patients can be distinguished from the status of those with other types of operations for the first two years following the pneumonectomy.

Comparison of (*a*) pneumonectomy completed after a thoracoplasty, (*b*) thoracoplasty performed after pneumonectomy to fill space, or (*c*) pneumonectomy completed without thoracoplasty, in the categories of active disease, inactive disease, and dead of tuberculosis does not reveal a significant advantage for any one type of operation over another.

Discussion

Eighty-eight per cent of these patients treated by pneumonectomy had far advanced pulmonary tuberculosis on admission to the sanatorium (2). All had disease in the unoperated lung, 39 per cent having at least moderately advanced disease in this lung. It is encouraging that 60 per cent of these patients had inactive disease on the first anniversary of their surgery, and that on the second anniversary 70 per cent had inactive disease (figure 1).

The possible advantages of thoracoplasty accompanying resection of the lung have frequently been discussed. There has been some sentiment expressed for concomitant thoracoplasty to reduce the overdistention of the opposite lung, particularly in patients with contralateral disease (3). Cruikshank and Papamichael (4) believed that a concomitant thoracoplasty should be performed to minimize the incidence of bronchopleural fistula and empyema. Bjork (5), in 1956, still recommended simultaneous osteoplastic wall thoracoplasty and pneumonectomy if the patient could tolerate the double procedure. Jones and Robinson (6), on the other hand, did not find a significant difference in complications between patients with and with-

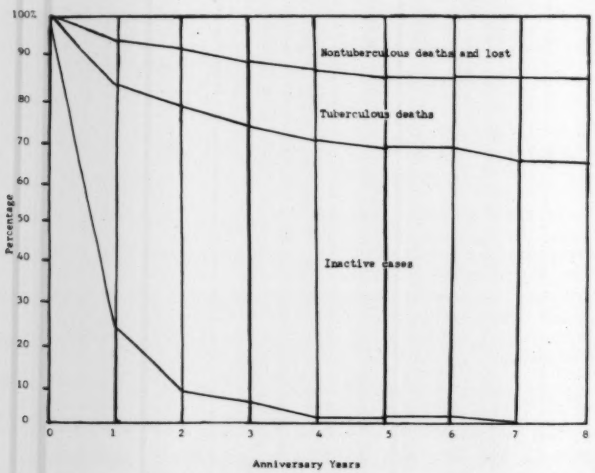

Fig. 1. Graphic presentation, using the anniversary method, of expected percentages in 204 pneumonectomy patients at the Firland Sanatorium from 1946–1955.

out postresection thoracoplasty. In the present series, the percentage of patients with inactive disease, active disease, or dead of tuberculosis is not significantly different when the previous thoracoplasty or concomitant thoracoplasty groups are compared. Although small in number, and with shorter follow-up examinations, the pneumonectomy without thoracoplasty group has similar values in these categories for the first two years. This is, however, the group that was treated more adequately with drugs.

Continuing activity of the disease or relapse is confined mainly to the first three years following pneumonectomy (figure 1). After this time only 2 per cent will have active pulmonary tuberculosis. This steady decline in the number of patients with active disease has been demonstrated by others for all classifications of tuberculosis (7–9).

When the results of treating tuberculosis with pneumonectomy are compared with the study of patients treated with thoracoplasty (10), it is evident that the disease is inactive in more pneumonectomy patients by the first anniversary date (60 per cent versus 49 per cent). After the third anniversary date a greater number of thoracoplasty patients (73 per cent) can be expected to have inactive disease (73 per cent versus 68 per cent). A greater percentage of active cases (18 per cent versus 5 per cent) is noted in the thoracoplasty group at this anniversary. Tuberculous deaths at the first anniversary in the pneumonectomy cases are expected to be higher (11 per cent versus 4 per cent), but by the eighth anniversary pneumonectomy results in only a slightly greater death rate (21 per cent versus 16 per cent). Some of the differences seen are undoubtedly explained by the fact that thoracoplasties were performed for less severe disease. Of the thoracoplasty group as a whole, 21 per cent had no contralateral disease, whereas all members of the pneumonectomy group had some contralateral disease at the time of admission to the sanatorium.

When the after-history of moderately advanced pulmonary tuberculosis (8) is compared with that of the patients undergoing pneumonectomy in this study, the former indicates a smaller incidence of death and higher incidence of active disease at each anniversary. When compared with patients having far advanced pulmonary tuberculosis treated only by sanatorium care, the patients treated with pneumonectomy have a much better prognosis (9).

Of some concern to those interested in control of tuberculosis is the number of patients lost to follow-up care. In this particular group, the number lost increased at a fixed rate of 3 per cent a year for the first two anniversaries, and then dropped off to between 1 and 2 per cent for the succeeding three years (figure 1). After the fifth anniversary, those being studied appeared to be constant in their attendance at the follow-up agency. That this number of patients lost to follow-up study is not excessive has been demonstrated by others who experienced a similar or greater incidence (7, 8, 11).

Summary

Two hundred and four patients who had pneumonectomy during the treatment of pulmonary tuberculosis from 1946 to 1955 were studied. The anniversary method was used to evaluate the outcome of these cases. At the first anniversary after the pneumonectomy, the patient has a 60 per cent chance of having inactive disease, a 24 per cent chance of having active disease. Eleven per cent will be dead on this anniversary of their pneumonectomy. At the eighth anniversary of the surgery, 63 per cent will have inactive disease, none will have active disease, and 21 per cent will be dead of their tuberculosis or from causes related to the pneumonectomy. The outcome of the pneumonectomy is not influenced by a previous or a concomitant thoracoplasty.

Sumario

La Neumonectomía en el Tratamiento de La Tuberculosis Pulmonar. La Historia Subsiguiente

Se estudiaron 204 enfermos a quienes se ejecutó la neumonectomía durante el tratamiento por tuberculosis pulmonar de 1946 a 1955. Se usó el método del aniversario para justipreciar el resultado en estos casos. En el primer aniversario de la neumonectomía, el enfermo cuenta con 60 por ciento de probabilidades de tener enfermedad inactiva, 24 por ciento de probabilidades de tener enfermedad activa. Un 11 por ciento habrá fallecido para dicho aniversario de la neumonectomía. En el octavo cumpleaños de la operación, 63 por ciento tendrán enfermedad inactiva, ninguno tendrá enfermedad activa y 21 por ciento habrán fallecido de la tuberculosis o de causas relacionadas con la neumonectomía. El resultado de la

neumonectomía no se ve afectado por una toracoplastia anterior o concomitante.

Resume

La pneumonectomie dans le traitement de la tuberculose pulmonaire. Les suites

L'étude porte sur deux cent quatre malades ayant subi une pneumonectomie au cours de leur traitement pour tuberculose pulmonaire, de 1945 a 1955. La méthode d'anniversaire a été utilisée pour evaluer les suites dans ces cas. Un an après la pneumonectomie, le malade a 60% de chance d'avoir une forme inactive de la maladie et 24% de chance d'avoir une forme active, 11% seront décédés pour le premier anniversaire de leur pneumonectomie. Au huitième anniversaire de l'opération, 63% auront une forme inactive, aucun n'aura de forme active et 21% seront morts de tuberculose ou de causes relatives à la pneumonectomie. Les suites de la pneumonectomie ne sont pas influencées par une précédente ou concomittente thoracoplastie.

Acknowledgment

The writers are indebted to Dr. Blair Bennett and Mr. R. L. Lessel for statistical assistance.

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THE TUBERCULIN TEST AND THE DIAGNOSIS OF CLINICAL TUBERCULOSIS[1-3]

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(Received for publication June 8, 1959)

Introduction

The disadvantage of excessive sensitivity to tuberculin was demonstrated in experimental animals by Rich (1) more than thirty years ago. However, there was no clinical support for this theory until about five years ago when Palmer and his associates showed that apparently healthy persons who have reactions of 10 mm. or more to a small dose of tuberculin, such as 0.0001 mg. of PPD (5 TU), develop clinical tuberculosis at a rate three to four times that shown by persons who have reactions of only 4 to 9 mm. to the same small dose. This significant difference was found first in the final analysis of the student nurse study (2) and in the study on Navy recruits (3), and has been confirmed by the British Medical Research Council (4). It can be accepted that the more severe tuberculin reactions occur in a specific group of the general population which has the maximal risk of developing clinical disease. These studies do not exclude the possibility, however, that the subjects with the 4 to 9 mm. reaction had formerly been infected with human tubercle bacilli.

The use of the tuberculin test as an aid in the differential diagnosis of patients who have symptoms or signs of illness is more complex than its use in surveys of apparently healthy subjects. Will practically all patients with clinical tuberculosis have positive reactions to 1:1,000 dilution of a potent preparation of Old Tuberculin (OT) or to 0.0001 mg. of PPD (5 TU), or must the strong doses such as 1:100 of OT or 0.005 mg. or second-strength PPD be used when the patient fails to react to the smaller doses? If the patient reacts only to the strong doses, is the reaction due to chemical irritation to the tuberculin itself, to a cross reaction from an infection with an atypical acid-fast organism, or is it caused by an infection with human tubercle bacilli?

A review of the literature reveals a difference of opinion concerning the significance of a single small dose of tuberculin. In 1937, Paretzky reported (5) that 316 (94.61 per cent) of 334 patients with active tuberculosis had positive reactions to 0.1 mg. or less of OT. In 1942, Furcolow, Hewell, and Nelson (6) found that 99.6 per cent of 468 patients with proved tuberculosis had reactions of 5 mm. or more to 0.0001 mg. or less of PPD. This latter figure of 99.6 per cent has been quoted widely in the literature, including an editorial in the *Journal of the American Medical Association* (7), as proof that only one dose of tuberculin is needed for clinical diagnostic studies. This conclusion was contrary to the clinical experience of the present writers, who proceeded to make a retrospective study of patients with tuberculosis in the Durham Veterans Administration Hospital (where PPD was used routinely) and in the Duke Hospital (where OT was employed).

Materials and Methods

Tuberculin reactions among patients in the Veterans Administration Hospital, Durham, North Carolina: The Durham Veterans Administration Hospital is a general medical and neurologic hospital with one ward of 30 beds for the treatment of tuberculosis. About 60.5 per cent of the patients are white and 39.5 per cent, Negro. A good many of the patients have been transferred from other services in the hospital, to which they had been admitted before tuberculosis was suspected, and the remainder are direct admissions from private physicians, public health clinics, regional offices, and medical wards of other Veterans Administration hospitals.

Fresh solutions of PPD were prepared from tablets purchased from Merck, Sharpe and Dohme by the Veterans Administration outlet station. All patients included in this study were originally tested with first-strength PPD (1 TU) followed by intermediate (5 TU), and by second-strength

[1] From the Veterans Administration Hospital and Duke Hospital, Durham, North Carolina.

[2] Presented before the Medical Session, as part of *Section 4C*, at the annual meeting of the American Trudeau Society, Chicago, Illinois, May 26, 1959.

[3] This investigation was supported, in part, by research grant E-1991 from the National Institute of Allergy and Infectious Diseases of the National Institutes of Health, U. S. Public Health Service.

PPD, if necessary, to obtain a reaction. All tests were given within five to fifteen days after admission.

Observations

Among 81 patients admitted consecutively, later proved by culture, biopsy, or necropsy to have tuberculosis, 68 (84 per cent) had reactions to first-strength and 76 (93.9 per cent) to intermediate-strength PPD. Five (6.1 per cent) failed to react to 5 TU. One was a nonreactor to second-strength PPD.

The types of cases studied are shown in table 1. There were 15 patients, not included in the series, in whom the dose of tuberculin was increased directly from first to second strength.

Tuberculin reactions among patients in the Duke Hospital: Except for certain patients admitted to the thoracic service, Duke Hospital does not encourage admission or knowingly admit patients with tuberculosis. The patients admitted for thoracic surgery have proved tuberculosis, but skin tests are not repeated and only a few are included in this series. In effect, the majority of the patients studied in this series were admitted to the medical and pediatric wards as diagnostic problems. These are the types of acutely ill patients who would be admitted to any large general hospital and, usually, would be more acutely ill than those admitted to Veterans Administration hospitals or to state sanatoriums.

Old Tuberculin has always been used in the Duke Hospital although checks with PPD are often made when a patient, suspected of having tuberculosis, fails to react to 1:100 OT. Three types of OT were used during the period covered by this study. The OT made in Saranac Lake was used for the earlier cases. For a short period, Lederle's OT was employed, but for the past eight years Wyeth's OT has been used exclusively. Aronson and associates (8) made direct comparison on the same subjects with PPD commercial, PPD-S, Saranac Lake, Wyeth, and other commercial tuberculin. The Saranac Lake tuberculin was definitely more reactive than PPD, and Wyeth's was in all respects equal to PPD. The present writers have made simultaneous tests with the Wyeth tuberculin and PPD in the medical students. With two different classes, different batches of Wyeth's tuberculin were compared with commercial PPD. The third class was compared with the standard PPD-S. In all three instances the results obtained with 1:100 of Wyeth was comparable to the second-strength tuberculin.

Most of the readings were done with the old "plus" system, but later measurement showed that, in general, reactions were being over-read rather than under-read. Reactions of 4 to 10 mm. were being considered 1-plus. The routine testing and reading were done by the medical students and house officers. They have always been given special instruction concerning the injection and the reading. The stock tuberculins are kept in the Department of Microbiology, and all dilutions are made and distributed by one experienced person. Failure to react cannot be attributed to obsolescent solution. Patients who failed to react to 1:100 OT were usually retested with a new, immediately fresh solution or with second-strength PPD or both.

The results obtained in 120 cases are shown in table 2. Many of the patients with proved tuberculosis did not have the complete series of skin tests, and many of the patients had the skin tests but lacked definitive proof of tuberculous infection. Hence, 120 cases are reported of patients with tuberculosis determined at necropsy or by microscopy, culture, biopsy, or guinea pig inoculation of respiratory secretions who also

TABLE 1

Skin Reactions to Tuberculin by Tuberculous Patients at the Veterans Administration Hospital, Durham, North Carolina

	Total	Definite Skin Reaction to			
		First-Strength PPD	Intermediate PPD	Second-Strength PPD	None
Extrapulmonary tuberculosis	11	9 (82%)	2 (18.0%)		
Far advanced tuberculosis	24	18 (75%)	4 (17.0%)	1 (4.2%)	1 (4.2%)
Moderately advanced tuberculosis	33	30 (91%)	1 (3.0%)	2 (6.1%)	
Minimal tuberculosis	11	10 (91%)	1 (9.1%)		
Miliary tuberculosis	2	1		1	
Total	81	68 (84%)	8 (9.9%)	4 (4.9%)	1 (1.2%)

When figured by cumulative reactions, there was a total of 5 patients who failed to react to 5 TU of PPD, giving a percentage of 6.1.

TABLE 2

SKIN REACTIONS TO TUBERCULIN BY TUBERCULOUS PATIENTS AT DUKE HOSPITAL, DURHAM, NORTH CAROLINA

Patients	Old Tuberculin 1:10,000		Old Tuberculin 1:1,000		Old Tuberculin 1:100	
	Reactors	Non-reactors	Reactors	Non-reactors	Reactors	Non-reactors
120	36 (30%)	84 (70%)	82 (69%)	38 (31%)	102 (85%)	18 (15%)

TABLE 3

SKIN REACTIONS TO TUBERCULIN BY TUBERCULOUS PATIENTS AT DUKE HOSPITAL, DURHAM, NORTH CAROLINA

Patients	Old Tuberculin 1:10,000		Old Tuberculin 1:1,000		Old Tuberculin 1:100	
	Reactors	Nonreactors	Reactors	Nonreactors	Reactors	Nonreactors
94	27 (29%)	67 (71%)	62 (66%)	32 (34%)	79 (84%)	15 (16%)

had the full series of skin tests. Among the 120 patients, 82 (69 per cent) reacted to 1:1,000 or more diluted solution of OT, while 38 (31 per cent) failed to react to 1:1,000, and 18 (15 per cent) failed to react to the 1:100 strength of OT.

The tuberculin test is applied as a routine in almost all problem cases on the medical and pediatric wards. The advantages of such a procedure are shown by the observation that tuberculosis was not considered in the differential diagnosis of 20 cases until the tuberculin test was found to be positive.

Among the 120 patients, 26 were diagnosed by microscopy and the diagnosis was not confirmed by sputum culture. To avoid the justifiable criticism that these cases may have contained a preponderance of atypical acid-fast infections and thus have inflated the percentage of patients who failed to react to 1:1,000 OT, they were eliminated from table 3. The percentage who failed to react to 1:1,000 OT was 34 and the percentage negative to 1:100 of OT was 16.

False positive reactions from chemical irritation of tuberculin: In the original standardization of PPD, Long and associates (9) found that 0.00002 mg. gave a reaction of approximately the same size in man as the 1:10,000 dilution of a good preparation of OT, and 0.005 mg. (second-strength) was the same as, or a little stronger than, a 1:100 dilution of OT. False reactions from chemical irritation from the 0.005 mg. dose were not observed. Later the 0.005 mg. dose was administered to a group of adult Negroes in a remote area of Dutch Guiana (10) and all failed to react except two who had been in contact with civilization.

A group of 8 Duke medical students, who gave a zero reaction to 0.005 mg. of PPD, volunteered to take a double dose, or 0.01 mg. Not one of the 8 volunteers had the slightest reaction. Six of the students were persuaded to take ten times the second-strength PPD, or 0.05 mg.

This dose did produce a chemical irritation which was quite marked in six hours, reached its maximum before twenty-four hours, and had practically disappeared by forty-eight hours. These observations in this small group of students would suggest that, when chemical irritation does occur from grossly excessive doses of PPD, the reactions begin before the sixth hour and can be readily differentiated from true allergic responses.

Ten milligrams of OT (1:10 dilution) were administered to more than 100 normal stock guinea pigs and rabbits without a false positive reaction to either the tuberculin or its corresponding control. These same animals gave specific tuberculin reactions to 10 mg. within seven to twenty-one days after experimental infection with tubercle bacilli. The degree of sensitivity gradually increased with time until most animals reacted to 0.1 mg. and some to 0.01 mg. of OT. In no instance was a positive reaction observed to the corresponding dilution of the control. Pediatricians frequently employ the 10-mg. dose in patients with miliary tuberculosis and meningitis. The use of the 10-mg. dose in adults was recommended by a number of clinical investigators in the 1930's, and it should be noted that Paretsky (5) who obtained positive reactions in 94.6 per cent of his 334 patients with tuberculosis recommended supplementary doses of 1 mg. and 10 mg. for the nonreactors to 0.1 mg. The patients of twelve additional cases, or 3.6 per cent, were found to be reactors.

Cross reactions in patients infected with atypical acid-fast infections: The reactions observed in man to second-strength PPD have all of the

characteristics of true allergic reactions of the delayed type, as to time or appearance, mode of evolution, character of induration, and degree of persistence. From inspection alone, a tuberculin reaction induced by 0.00002 of PPD or 0.005 of PPD cannot be differentiated from an allergic reaction induced by histoplasmin or coccidioidin. Fortunately, there are no cross reactions between these fungal antigens and tuberculin. But there are cross reactions between tuberculins made from human tubercle bacilli and tuberculins made from a great variety of pathogenic and nonpathogenic species of mycobacteria. Patients infected with atypical acid-fast organisms may have 10 mm. or more of induration to 0.0001 mg. of PPD (5 TU), but usually react only to 0.005 mg. or second-strength (11) PPD. But some patients infected with human tubercle bacilli fail to react to 5 TU but do give reactions with second-strength PPD.

What is the clinical significance of a reaction to second-strength PPD? There is a mass of statistical data which suggests that most of these reactions to strong tuberculin are cross-allergic reactions induced by subclinical infections with atypical acid-fast bacilli or other organisms capable of inducing allergy to tuberculin (12). There are others who believe that these are true tuberculin reactions to minimal and to obsolescent subclinical infections with true tubercle bacilli. There is no adequate statistical support for this alternate theory. But there is conclusive statistical data from the British study (4) and from the student nurse study (2) that subjects reacting only to strong doses of tuberculin have a relative immunity to clinical tuberculosis which is two to three times that of those with 10 mm. or more of induration to 5 TU and those who are nonreactors to second-strength PPD. Second-strength PPD thus differentiates those who have never been infected, and are the most susceptible to disease when infected, from the second-strength reactors who are the most resistant group.

Discussion

The percentage of patients with clinical tuberculosis who will react to 0.0001 mg. of PPD (5 TU) or to 1:1,000 of OT varies from hospital to hospital and depends apparently on a number of factors such as race, extent and type of disease, and the degree of illness at the time when the test was performed (13, 14).

Pepys (15) has reviewed the physiologic changes which will increase or decrease the size of the tuberculin reaction but, if the patient with clinical tuberculosis is in good physical condition, his skin of normal tone, and is not taking corticosteroid hormones, then more than 99 per cent will have reactions to 5 TU of PPD. Mascher (16) was able to collect 11 cases of tuberculous patients who were in apparently good condition but failed to react either to small or large doses of tuberculin, and the present writers have observed several similar patients. The number of such cases is so small, however, that it does not seriously invalidate the diagnostic usefulness of tuberculoprotein.

Another factor comes into play when dealing with sick patients. The physiologic tone of the skin seems to be altered, so the true degree of allergy is not revealed by the skin test. Some patients with chronic progressive pulmonary disease, tuberculosis of the pleura, pericardium, and peritoneum, or with overwhelming infection such as miliary tuberculosis or meningitis, show a progressive decrease in allergy which may result finally in failure to react to tuberculin in any concentration. It would be logical to assume that among a large group of definitely sick but not moribund patients there would be many more who could react to second-strength PPD than to 5 TU.

In a well-fed and stabilized sanatorium population, Furcolow, Hewell, and Nelson (6) found that the 5-TU dose of tuberculin gave reactions in 99.6 per cent of 468 proved cases of tuberculosis. It was observed, however, that the sicker patients gave smaller reactions than the apparently healthy. Lester and Atwell (17) extended this type of study by performing the tuberculin tests on admission to the sanatorium, at which time more critically ill patients would be encountered. Among 624 patients subsequently found to have tuberculosis, 50 (8.7 per cent) failed to react to 5 TU, and 4 did not react to second-strength. However, all 4 subsequently had satisfactory tuberculin reactions after their physical conditions were improved by therapy. Among 304 soldiers admitted to Fitzsimons (18), 9 (2.3 per cent) failed to react to 5 TU and 4 (1.3 per cent) did not react to second-strength.

Twelve years after the original study by Furcolow, Hewell, and Nelson (6), Hewell and

Suyemoto (19) restudied the patients in the same sanatorium and found that only 95 per cent, in contrast to 99.6 per cent, reacted to 5 TU. But 17 (8 per cent) among 211 white male patients did not react to the 5-TU dose. Standard PPD-S was used in both studies, and Hewell was an active investigator in both. It must be assumed that, by chance, there was a larger proportion of severely ill patients in the hospital during the second study than during the first.

The 6.1 per cent of tuberculous patients in the Durham Veterans Administration Hospital who failed to react to 5 TU on admission is somewhat smaller than the 8.7 per cent found in a state sanatorium in Ohio by Lester and Atwell.

The number of patients in the Duke Hospital who failed to react to 1:1,000 OT was surprising. The expectation was that more than 5 per cent but less than 10 per cent would fail to react. The tuberculin was potent, the dilutions were accurate, and the solutions fresh. Reactions as small as 4 mm. were read as positive. A general hospital population was being studied, in which there would be more acutely ill patients than in either a Veterans Administration hospital or a state sanatorium. Thirty-eight of the 120 were more than fifty years of age. Many of the patients had generalized, but not typical, miliary tuberculosis.

The diagnostic difficulties are shown by a review of the last 100 consecutive necropsies at Duke Hospital showing active tuberculosis. Of these 100, 21 persons were not suspected of having tuberculosis before necropsy. The results from the Herman Kiefer hospital in Detroit are more nearly comparable with those at Duke since both hospitals have a large percentage of Negroes and low-income white patients. However, more less severely ill patients would be admitted to Herman Kiefer since ambulatory patients found to have tuberculosis are not admitted to Duke. Musacchio reported in 1940 (20) that the 1:1,000 (0.1 mg.) dose of OT missed 10 (15 per cent) of 67 minimal cases, 37 (10.9 per cent) of the 340 moderately advanced, and 80 (15.5 per cent) of the 511 far advanced cases.

A confirmation of Masacchio's results in far advanced cases can be found in the report of Salomon and Angel (21) who tested 63 patients with far advanced disease with 5 TU of PPD before they were randomized for drug therapy with and without corticotropin. Twelve of the 63 (19 per cent) were nonreactors to this dose although all were reactors at the end of the test after corticotropin was discontinued.

One variant in the present study, in both the Durham Veterans Administration Hospital and in the Duke Hospital, was the fact that the tuberculin tests were administered and read by many different persons and not by a single individual or a single team. The percentage of negative reactions to the 5-TU dose was less in the Durham Veterans Administration Hospital than in the state sanatorium studied by Lester and Atwell (17), where the single-team study was employed. Even more subjects were involved in the Duke Hospital study than in the Durham Veterans Administration study, and the percentage of negative reactions was higher than in any previously reported series. It is not practical in a general hospital to have all of the tuberculin tests performed and read by a single individual or a single team. The frequency of negative reaction to 1:1,000 OT in Duke Hospital illustrated the importance of always giving 1:100 OT or second-strength PPD when the reaction to 1:1,000 or 5 TU is doubtful or negative.

Summary

Reactions to 1 mg. of OT or to second-strength PPD (0.005 mg.) are true allergic reactions and not the result of chemical irritation from tuberculin. The allergy may be from an atypical acid-fast organism or from a true tubercle bacillus.

A negative reaction to 1 mg. of OT or 0.005 mg. of PPD by patients who are in reasonably good physical condition almost eliminates the possibility of infection with either atypical acid-fast organisms or human tubercle bacilli. Healthy subjects reacting only to second-strength PPD (0.005) are more resistant to the development of clinical tuberculosis than those who react strongly to 5 TU and those who fail to react at all to second-strength PPD. Relative immunity is assured by this low-grade allergy regardless of whether it is induced by atypical acid-fast organisms, other unknown organisms, or true tubercle bacilli.

The percentage of patients with clinical tuberculosis who failed to react positively to intermediate doses of tuberculin (5 TU) varied from a low of 0.4 per cent in one state sanatorium

to 5 per cent in the same sanatorium twelve years later; to 6.1 per cent in a Veterans Administration hospital, to 8.7 per cent in a different state sanatorium, and to 31 per cent in a general hospital.

Sumario

La Prueba de la Tuberculina y el Diagnóstico de la Tuberculosis Clínica

Las reacciones a 1 mg. de TA o a la segunda concentración (0.005 mg.) de PPD (DPP) son verdaderas reacciones alérgicas y no la consecuencia de irritación química producida por la tuberculina. La alergia puede proceder de un microbio ácidorresistente atípico o de un verdadero bacilo tuberculoso.

Una reacción negativa a 1 mg. de TA o a 0.005 mg. de PPD por enfermos que se hallan en estado físico bastante bueno casi elimina la posibilidad de infección por ya microbios ácidorresistente atípicos o bacilos tuberculosos humanos.

Los sujetos sanos que no reaccionan más que a la segunda concentración (0.005 mg.) de PPD son más resistentes a la aparición de tuberculosis clínica que los que reaccionan intensamente a 5 U.T. y los que no reaccionan en lo más mínimo a la segunda concentración de PPD. Con esa débil alergia se asegura inmunidad relativa, independientemente de si la provocan atípicos microbios ácidorresistentes, otros microbios desconocidos o verdaderos bacilos tuberculosos.

El porcentaje de enfermos con tuberculosis clínica que no reaccionaron positivamente a dosis intermedias de tuberculina (5 U.T.) varió de un fondo bajo de 0.4 en un sanatorio público a 5 en el mismo establecimiento doce años después; a 6.1 en un hospital de la Administración de Veteranos, a 8.7 en otro sanatorio público y a 31 en un hospital general.

Resume

Le test à la tuberculine et le diagnostic clinique de tuberculose

Les réactions obtenues avec un milligramme de tuberculine vieillie ou avec 0,005 mg de tuberculine purifiée sont de vraies réactions allergiques et non le résultat d'une irritation chimique produite par la tuberculine. L'allergie peut venir d'un germe acido-resistant ou d'un vrai bacille tuberculeux humain.

Une réaction negative à un milligramme de tuberculine vieillie ou à 0,005 mg de tuberculine purifiée, chez des malades qui sont en à peu près bonne condition physique elimine presque définitivement la possibilité d'une infection par un germe acido-résistant atypique ou un bacille tuberculeux humain.

Des sujets en bonne santé réagissant seulement à 0,005 mg de tuberculine purifiee sont plus résistants au developpement d'une forme clinique que ceux qui reagissent fortement à 5 unités de tuberculine et que ceux qui ne réagissent pas du tout. Une immunité relative est assurée par cette allergie mineure pu'elle soit provoquée par des germes acido-resistant typiques, par d'autres germes inconnus ou par de vrais bacilles tuberculeux.

Le pourcentage de malades avec une tuberculose clinique, qui ne réagit pas d'une façon positive à 5 unités de tuberculine (dosage moyen), varie entre 0,4% dans un sanatorium d'Etat, et 5% dans le même sanatorium 12 ans plus tard; 6,1% à l'hopital de l'administration des Vétérans; 8,7% dans un autre hopital d'Etat et 31% dans un hopital général.

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INTERMEDIATE-STRENGTH PURIFIED PROTEIN DERIVATIVE

Its Diagnostic Significance in Active Tuberculosis[1,2]

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(Received for publication June 19, 1959)

Introduction

It has been conceded for a long time that tuberculin skin testing is one of the most powerful aids in the diagnosis of tuberculosis. It attains greater significance with each passing year, with an ever-increasing tuberculin-negative population. Since the excellent work of Palmer (1) has convincingly shown that the second-strength purified protein derivative (PPD) skin test has limited value because fully 33 per cent of the population will give nonspecific reactions, it has been apparent that the true significance of tuberculin skin testing hinges on the validity of the reaction to intermediate-strength PPD.

Reports have lately appeared stating that the intermediate-strength PPD skin reaction will be negative in a significant number of active cases of tuberculosis, enough to limit seriously the diagnostic value of the test. As early as 1942, Furcolow, Hewell, and Nelson (2) reported a positive skin reaction to an intermediate dose of a standard PPD preparation in 99.6 per cent of 468 tuberculous patients. Other workers (3–7), however, have not attained this percentage. Lester and Atwell (3) recently stated that the reaction to intermediate PPD was negative in 8 per cent of their patients with tuberculosis. In an effort to bring into clearer focus the true significance of this test, the present writers undertook a critical reappraisal of its specificity in active tuberculosis, in a two-part, prospective study.

Material and Methods

Four hundred and sixteen patients with tuberculosis, proved either bacteriologically and/or histologically, were studied. The skin reaction to commercially available purified protein derivative intermediate (0.0001 mg.) antigen[4] was determined. A positive skin reaction was defined as 5 mm. of induration or greater at the site of intradermal injection within forty-eight to seventy-two hours. The majority of cases, 78 per cent, were classified as far advanced; 20 per cent, moderately advanced; and 2 per cent were classified as minimal (table 1). Males predominated in the series. Cultures were positive for tubercle bacilli in 95.4 per cent of the patients, and the remaining 4.6 per cent had caseating granulomas, frequently with cultures positive for tubercle bacilli. Thus, the disease in the 416 patients proved to be active tuberculosis. The study was designed so that the skin test with intermediate-strength PPD was routinely repeated whenever the reaction was initially negative or read as a doubtful reaction.

Results

As may be seen in figure 1, 386 patients (92.8 per cent of the total) gave positive reactions the first time they were studied. Thirty patients (7.2 per cent) were negative, however, to the initial test; but when the same intermediate dose of PPD was reapplied, all but 7 patients (1.7 per cent of the total) had positive reactions.

When this group of 7 patients who, with proved tuberculosis, had negative reactions to two successive PPD-intermediate skin tests was analyzed, it was found that all but one patient had very obvious cause for anergy. Three patients had far advanced tuberculous disease, were moribund, and eventually died. One patient with moderately advanced tuberculous disease was extremely toxic from a gangrenous extremity at the time of his skin tests. Another patient had miliary tuberculosis and was receiving steroid therapy from the onset, which explained his anergy. It is interesting to note that all 5 of these patients were more than sixty-five years of age and that death was the outcome in 3 of them. The life of the fourth is presently hanging in delicate balance. Another patient, the only female, had proved sarcoidosis and then developed tubercu-

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[4] Manufactured by Merck, Sharp and Dohme.

TABLE 1

ANALYSIS OF SEVEN PATIENTS REPEATEDLY NEGATIVE TO INTERMEDIATE PPD

Case	Condition	Age	Color	Sex	Outcome
1	FA, moribund	70	Nonwhite	M	Deceased
2	FA, moribund	70	White	M	Deceased
3	FA, moribund	65	White	M	Deceased
4	MA, very toxic	78	Nonwhite	M	Moribund
5	Miliary tuberculosis during corticosteroid therapy	65	Nonwhite	M	Well
6	Tuberculosis and sarcoidosis	40	Nonwhite	F	Well
7	MA sickle cell anemia (no apparent cause of anergy)	45	Nonwhite	M	Well

FA—far advanced
MA—moderately advanced

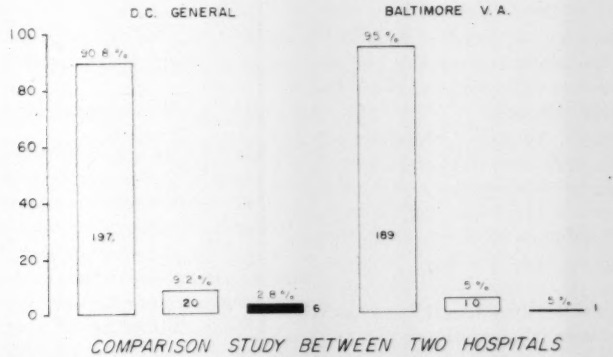

FIG. 1. Comparison of the results of skin testing with intermediate-strength PPD at two hospitals. When the 6 patients who were repeatedly negative in the District of Columbia General Hospital group are excluded because of anergy, there is striking similarity between the two groups.

losis. Her skin reactions have been negative while she has been receiving intermittent steroid therapy. Only one patient had no obvious and striking cause for anergy. He did have a skin reaction but, by the present criteria, it could only be classified as doubtful, even when repeated.

The true significance, then, of the intermediate dose of PPD lies in the fact that it is 99.76 per cent accurate, with a false negative incidence of only 0.24 per cent when the skin reaction is restudied in all of the patients with initially negative or doubtful reactions and when patients with obvious cause for anergy were excluded.

DISCUSSION

From these data, it may be seen that it is the repetition of the skin test in those patients with initially negative or doubtful reactions that is of prime importance. This fact has been recognized by Wier and others (5, 8) but has not been sufficiently stressed. The present writers believe that most of the reports in the literature that claim that the intermediate PPD skin test is positive in 90 to 98 per cent of patients with active tuberculosis have been based on retrospective analyses of hospital charts. In such analyses, no allowance is made for the errors in technique in the administration of an intradermal test, errors that are so many and so well known they hardly need further elucidation here. These are errors, however, that can conceivably affect the false negative percentage by as much as 6 per cent. Another factor which is well covered by repeating the initially negative skin test is that of transient tissue anergy. The vast majority of the 23 patients of this study who had initially negative reactions and then had positive ones when retested were not elderly and extremely dehydrated, as is true in the classical description of the anergic patient. A surprising number were febrile, but in no sense

of the term could they be considered as being critically ill. Based on this study then, included by the present writers in the list of obvious causes of anergy are: only the extremely ill, moribund patient; the patient with miliary tuberculosis; the patient with sarcoidosis; the patient receiving steroid therapy; and the patient with protracted remittent fever.

To demonstrate that the present results were reproducible in the clinical setting of every-day hospital practice, the study was divided between two hospitals. At the District of Columbia General Hospital, all of the skin tests were applied by one of three investigators and each reaction was then read in triplicate, each investigator reading independently of the other. Unless all three readings were in agreement, the reaction was classified as doubtful and considered negative for purposes of our tabulation. This applied to both the initial and repeated skin tests.

At the Baltimore Veterans Administration Hospital, the skin tests were applied as part of the routine ward procedure by the house staff or by medical students and were read without triple checking. The procedure of repeating all negative or doubtful reactions was the same, however. A comparison of the results shows a striking similarity between the two hospitals. This is particularly true when it is considered that all 6 patients with negative reactions whose tests were repeated at the District of Columbia General Hospital can be excluded because of obvious causes of anergy. The higher incidence of initially negative reactors at the District of Columbia General Hospital can undoubtedly be attributed to the type of patient admitted to an active city hospital. Since the majority of patients were acutely ill on admission, they would be more apt to have an initially negative skin reaction because of toxicity than the referral type of patient seen at the Baltimore Veterans Administration Hospital.

Concerning the time interval between tests, an attempt was usually made to repeat them within a week, in an effort to establish early diagnosis. If a patient was toxic, however, and suspected of having a transient tissue anergy, retesting was frequently deferred until a change was noted in the clinical course. There are those who say the skin reaction will be positive or negative depending upon when in the course of his disease the patient is tested (6). While most of the patients of the present study were skin tested on and shortly after their admission, many (particularly at the Veterans Administration Hospital) had been hospitalized and had been receiving therapy for six to twelve months before the study was undertaken. The one instance in which it is believed that the time interval between skin tests is of the utmost importance is in the evaluation of a tuberculosis contact. Here the incubation period of twenty-two to sixty days must be kept in mind.

In conclusion, the present writers believe that, faced with any patient, regardless of how suggestive his roentgenographic or clinical course may be for tuberculosis, if the intermediate-strength PPD skin reaction is negative after two properly applied tests and no obvious cause for anergy (including toxicity) exists, then it is extremely unlikely that the patient has tuberculosis.

Summary

The value of the skin reaction to intermediate-strength PPD as a diagnostic tool in active tuberculosis is evaluated in a prospective study of 416 tuberculous patients. Of the 416 patients, 7.2 per cent were nonreactors to the initial test but, when it was repeated in these same patients, only 1.7 per cent were nonreactors.

Examination of the 1.7 per cent who were persistently negative to the intermediate-strength PPD revealed that all but 0.24 per cent had obvious cause for anergy, making the test 99.76 per cent accurate.

The factors contributing to initially negative skin reactions and the importance of repeating the test when the reaction is negative or doubtful are emphasized. Evidence is presented to show that these results are reproducible in the usual clinical setting of hospital practice.

Sumario

El PPD (DPP) Intermedio: Su Importancia Diagnóstica en la Tuberculosis Activa

Se determinó el valor de la cutirreacción con el PPD (DPP) intermedio como instrumento de diagnóstico en la tuberculosis activa en un estudio anticipado de 416 tuberculosos. De los 416, 7.2 por ciento resultaron negativos a la cutirreacción inicial, pero al repetirla en los mismos enfermos, sólo 1.7 por ciento fueron negativos.

El examen del grupo de 1.7 por ciento que fué persistentemente negativo al PPD intermedio reveló que todos, menos 0.24 por ciento, tenían

alguna causa manifiesta de la anergia, lo cual dota a la prueba de una exactitud de 99.76 por ciento.

Recálcanse los factores que contribuyeron a la negatividad inicial de las cutirreacciones y la importancia que reviste el repetirlas cuando son negativas o dudosas.

Ofrécense datos demostrativos de que estos resultados son reproducibles en el habitual cuadro clínico de la práctica hospitalaria.

Resume

Tuberculine purifiée. Sa signification diagnostique dans les formes actives de tuberculose

La valeur de la cuti-réaction avec la tuberculine purifiée, en tant que moyen de diagnostic dans la tuberculose active est etudiée chez 416 tuberculeux. Parmi ces malades, 7,2% avaient une cutiréaction négative, au début, mais, quand le test fut répété chez les mêmes malades, seulement 1,7% eurent une cuti-réaction négative.

L'examen des 1,7% qui persistèrent à être négatifs avec cette tuberculine montra que tous, excepté 0,24% avaient une raison évidente d'être anergiques, ce qui fait qu' en réalité, ce test est fidèle dans 99,76% des cas.

Les auteurs insistent sur l'étude des facteurs responsables d'une réponse initiale négative et l'importance de répèter le test dans les cas négatifs ou douteux. Il est evident que ces résultats peuvent être employés dans la pratique clinique courante à l'hopital.

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STUDIES ON THE INTERACTION BETWEEN HOST CELL AND PARASITE[1]

I. Morphologic Studies of Lung Tissue Cultured *in Vitro*

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(Received for publication December 29, 1958)

Introduction

Does the alveolar epithelium that covers the pulmonary alveolar wall really exist, or not? Are the so-called "dust cells" or "septal cells" or alveolar phagocytes actually derived from alveolar epithelium; or are they from the connective tissue? Histology has gone through many decades with little knowledge regarding these questions. Opinions have diverged widely. Recently, however, electron microscopic studies by Low and Daniels (1), Low (2), van Breemen and Neustein (3), and Karrer (4) are adding more weight to the argument that "there is a thin continuous cellular covering of the alveolar wall, and this thin cellular layer is distinguishable from the endothelium of the capillary."

More recently, Low and Sampaio (5) introduced colloidal thorium dioxide (ThO_2) into the pulmonary alveoli and observed at electron microscopic mangification the reaction of the alveolar epithelial cells and the alveolar phagocytes against this foreign material. They observed that, while the alveolar phagocytes were typically phagocytic, the alveolar epithelial cells did not ingest the foreign element. This led them to propose that the alveolar epithelial cells are of entodermal origin and the alveolar phagocytes are mesodermal. Maximow and Bloom (6) claimed that septal cells and dust cells were identical with the alveolar phagocytes, and were indistinguishable from those macrophages seen elsewhere. According to this concept, alveolar phagocytes do belong to the connective tissue and have little to do genetically with alveolar epithelial cells.

The present writer has cultured embryonic lung tissues of the guinea pig to see the interaction between tubercle bacilli and the cells of the lung. The results were generally in accordance with the recent electron microscopic findings, but different from the reports made by Lang (7–9) and Timofejewsky and Benewolenskaja (10). The present paper concerns the tissue culture technique and the morphologic aspect of the lung tissue cultured *in vitro*. The bacteriologic aspect of the same study will be presented in a separate paper.

Materials and Methods

Embryonic lung peripheries of guinea pigs, pregnant forty to fifty days, were used. Some tissues were used without the pleural covering.

A small Petri dish with an inner diameter of 27 mm. contained an 18-sq. mm. coverglass (figure 1). This coverglass was placed in the dish before the manipulation. For quick outgrowth of a minutely small fragment of tissue, tryptically digested, a supporter is of great value. The supporter was a coating over the coverglass of heparinized chicken plasma diluted three times with Tyrode's solution. This thin plasma film was prepared by pouring the diluted solution not only over the coverglass, but also over the entire bottom of the Petri dish, and then draining it off. Before the plasma dried, chick embryo extract, diluted ten times with Tyrode's solution, was poured over the wet coverglass and the Petri dish, and the residue was pipetted out completely. A clotted film was ready in ten to fifteen minutes, and the coverglass was fixed to the bottom of the Petri dish.

So that the outgrowth of the cells might be easier and quicker and so that the emigrated zone might be a monolayer, the section taken from the lung was digested with trypsin in the following way: A few pieces of tissue were placed in a short test tube, 1.5 cm. in diameter and 6 cm. in length, and were cut still finer with scissors. The blood was then washed away with Rinaldini's solution. Then three parts of a trypsin solution were added to each part of the washed tissue in the short test tube. This solution contained 1,000 H.U.M.[2] of crystalline trypsin per 1 ml. of Rinaldini's solution. The tube was placed in a water bath at 37°C. This tryptic digestion is not to single out a cell, but to loosen intercellular space and to obtain a still finer mass of the tissue. Care must be taken, therefore, in order that the cell will not be injured by the enzyme. Density of trypsin and duration of digestion should be adjusted to the development of the embryo. As the digestion proceeded, the tissue mass decomposed into a slimy

[1] From the Kitasato Institute for Infectious Diseases, Tokyo, Japan.

[2] The crystalline trypsin used is a product of Mochida Pharmaceuticals, Tokyo, and its H.U.M. (hemoglobin unit of Mochida) represents about 0.5 mg.

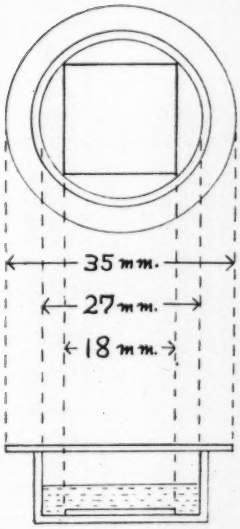

FIG. 1. Tissue culture chamber.

substance. Repeated pipetting left many minute tissue masses macroscopically visible fifteen to twenty minutes from the beginning of digestion.

The test tube was removed from the water bath, and a large quantity of Tyrode's solution that had been cooled was added. By this procedure the activity of the digester decreased abruptly. The tube was allowed to stand one to two minutes, permitting the undigested gross masses of tissue to sink. After the larger ones had settled, the upper fluid in the tube in which finer tissues were suspended was transferred to another tube. This tube was centrifuged for three minutes at 1,500 r.p.m. The sediment was resuspended for another washing in Tyrode's solution.

Completely washed minute masses of tissue were suspended in a nutrient fluid that was composed of 60 per cent of Tyrode's solution, 30 per cent of inactivated horse serum, 10 per cent of C.E.E.II (a 100 per cent extract from a nine-day chick embryo diluted with an equal amount of Tyrode's solution), and 50 units of penicillin per ml. of the medium. The pH of the medium was adjusted to 7.4–7.6 with 1 per cent sodium bicarbonate.

Then the suspension, 2 ml. each, was cultured. The culture chambers were then covered with the glass soaked in sterilized hot paraffin to prevent evaporation of the medium, followed by incubation at 37°C. The medium was made fresh every forty-eight hours.

Two cultures were taken every twenty-four hours for two weeks after culture, were fixed in 10 per cent neutral formalin for thirty minutes, and stained with Giemsa stain.

RESULTS

The results of tissue culture in the first twenty-four hours are shown in figure 2. The fine tissue masses were already giving squamous epithelial cell sheets, extending here and there like islands over the plasma film. Between the islands many spindle cells and macrophages were seen. When the cultures were two to three days old, the cell sheets spread much wider than in the one-day culture. When the tissue density of the suspension was high and the masses were fine, the entire culture surface was evenly covered with a monolayer of epithelial cells (figure 3). The islands often contained necrotic cells in their centers, if the masses of the tissue were not fine enough. The individual cells of the sheet were usually clearly separated from each other (figure 4). The protoplasm was very wide and tended to be wider as the culture progressed. The nucleus was round or oblong, in almost all cases one to each cell. Occasionally, however, one cell might have two to three nuclei. The nucleus stained lighter than that of macrophages, and similar to that of spindle cells. One or several nucleoli were seen rather clearly. The cells were stable in culture and did not give conspicuous changes even after two weeks of culture. Mitosis, however, was not seen in these cells.

Spindle cells occurred not only among the epithelial cell sheets, but also in the island itself, with macrophages forming strands and clumps. The strands and clumps were connected to form septa over the epithelial cell sheets. The whole picture looked like a net (figures 5, 6). The strands sometimes gave very long chains of parallel spindle cells. Unlike epithelial cells, these showed mitosis.

Macrophages are, at a glance, similar to individually isolated epithelial cells, but are apparently different from them in the following points. Macrophages, spreading protoplasm—sometimes with characteristic pseudopodia—adhered to the culture surface within a very short time after the tissue suspension was placed in the culture chamber. These cells isolated by tryptic digestion usually adhered to the coverglass within thirty minutes, spreading their protoplasm (figure 7). At this stage, epithelial and spindle cells did not spread their protoplasm on the coverglass, but macrophages seemed to be easily injured by trypsin digestion, because those macrophages that are thus isolated in the early stage of cultiva-

FIG. 2 (Left). Twenty-four hour culture. (80×) The fine tissue masses of lung are giving squamous epithelial cell sheets extending here and there like islands over the culture surface. Isolated cells between the sheets are mostly spindle cells and a few macrophages.

FIG. 3 (Center). Three-day culture. (80×) The entire culture surface is covered with a monolayer of the epithelial cell sheets. This is observable only when the tissue mass is extremely fine and the tissue density of the suspension is relatively high.

FIG. 4 (Right). Six-day culture. (600×) High-power magnification of the epithelial cell sheet. Cells are rather clearly separated from each other.

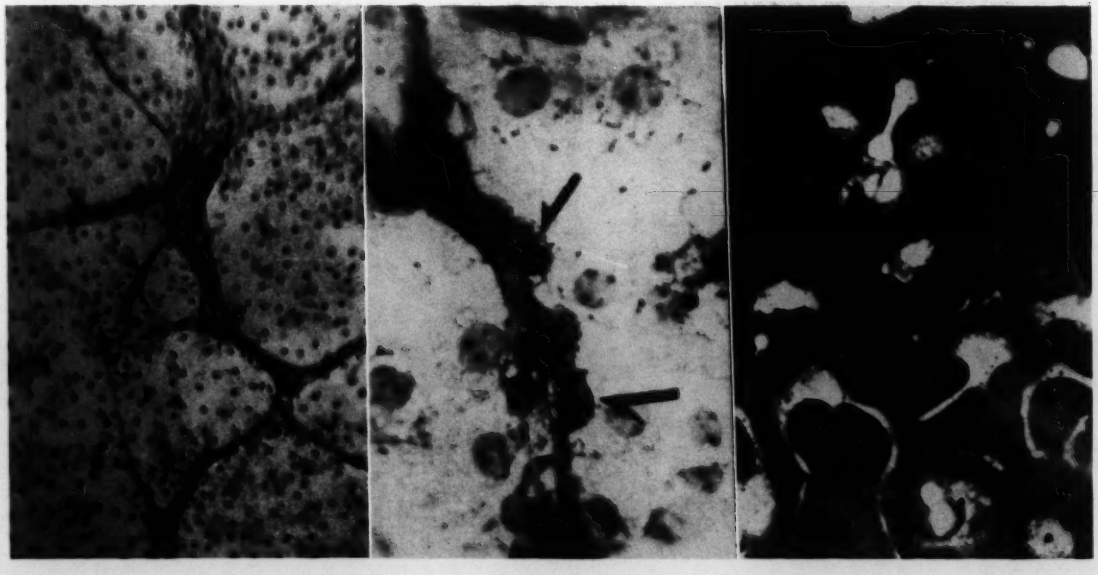

FIG. 5 (Left) Five-day culture. (120×) Strands and clumps made up of spindle cells and macrophages are connected into a mesh as if dividing the epithelial cell sheet with septa.

FIG. 6 (Center). Three-day culture. (800×) Macrophages seen attached to the cell strand (↑). The nucleus here usually stains darker than that of epithelial cells.

FIG. 7 (Right). Macrophages that are spreading their protoplasm adhere on the culture surface in a very short time. One hour culture; phase-contrast picture. (300×).

tion do not survive very long. Most of them degenerate within twenty-four to forty-eight hours. Most macrophages observable three to four days after cultivation appeared with spindle cells, forming cell clumps and strands. The nucleus stained darker than that of epithelial cells and spindle cells. The cell is the most unstable among the three kinds. It easily degenerates and disintegrates *in vitro*. Mitosis was not observed here either.

Discussion

Lang believed that the epithelial cells he saw in the lung tissue of adult rabbits cultured *in vitro* were derived from the bronchial epithelium. He denied the existence of alveolar wall epithelium except on nonnucleated plates (7–9). Timofejewsky and Benevolenskaja, on the contrary, concluded that the strongly phagocytizing cells that migrated in the initial stage of culture were phagocytes of alveolar epithelial origin (10). But these are the very cells that Lang called septal cells and that the present report classifies as macrophages. They are totally unlike the epithelial cells treated here.

Thus, former workers in the field of tissue culture could not have observed alveolar epithelial cell sheets or, if they could, paid little attention to them (11).

For the origin of the cells which formed the epithelial cells sheets as seen in this experiment, the following may be considered: pulmonary alveolar epithelial cells, bronchial epithelial cells, and the pleural cells. Origin from pleural cells is not likely because the results obtained from the tissue previously made free of pleura and from the tissue not treated in that way were almost identical. Differentiation of the bronchial epithelium from the alveolar epithelium as the origin of the sheet is the hardest to confirm; in fact, it is almost impossible to do so. The sheet, on the other hand, might have both. However, the present writer believes that the sheets were chiefly of alveolar epithelial origin for the following reasons: In this series of experiments, the tissues were all from the peripheries of the lung, and special care was taken not to let the bronchi be included. The sheets, however, grew out from each minute mass on the culture surface and, the finer the tissue mass got, the more the epithelial cells alone appeared. Again, the number and the area of epithelial cells were more than half of the emigrated cells. It was usual to see strands and clumps of the mixture of spindle cells and macrophages dividing the sheets. The writer believes that these cell strands and clumps are the connective-tissue part of the alveolar walls, because, under tissue culture conditions, the epithelial cells would be spreading into monolayers because of their specific nature, while the connective-tissue parts, separated at the basement membranes, would be left *in situ*.

As indicated in the results above, the macrophages are apparently different from the epithelial cells morphologically. Furthermore, they can easily be distinguished from other cells by their strong phagocytic property that has been reported in another experiment with tuberculous infection (12). This indicates that macrophages, alveolar phagocytes, are of connective-tissue origin as Low and Sampaio (5), Lang (7–9), Foot (13), and Gardner and Smith (14) believed.

The spindle cells seen in the cultured pulmonary tissue must be from capillary endothelium and from fibroblasts.

Summary

In culturing tiny lung tissue particles, loosened by trypsin treatment from the guinea pig embryo on a coverglass coated with the plasma film, an outgrowth was observed of epithelial cell sheets whose constituent was chiefly alveolar epithelial cells. Macrophages and spindle cells grew out on the same culture surface.

Sumario

Estudios de la Interacción entre las Células y los Parásitos del Huésped

Al cultivar partículas minúsculas de tejido pulmonar, desprendidas por el tratamiento con tripsina del embrión de cobayo sobre un cubreobjetos recubierto con una película de plasma, se observó una excrecencia formada por hojas de epiteliocitos cuyo componente consistía principalmente en células epiteliales alveolares. En la misma superficie del cultivo proliferaban macrófagos y células fusiformes.

Resume

Etude des réactions réciproques de la cellule hôte et du parasite

La culture de particules minuscules de tissu pulmonaire, traité par la trypsine de l'embryon de cobaye, pour obtenir un relâchement, sur un couvercle de boîte de culture enduit d'une pellicule de

plasma, permet d'observer la croissance de couches de cellules épitheliales composées essentiellement de cellules épithéliales alvéolaires. Des macrophages et des fibroblastes ont poussé sur la même culture.

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STUDIES ON THE INTERACTION BETWEEN HOST CELL AND PARASITE[1]

II. The Reaction of Pulmonary Cells Cultured *in Vitro* to Virulent Tubercle Bacilli

KEONG WUN LYANG

(Received for publication December 29, 1958)

Introduction

Since the quantitative technique has been applied to experimental tuberculosis in animals, it has become clear that the fate of tubercle bacilli differs from organ to organ. According to Pierce and associates (1), the number of living bacilli in the lung continues to increase throughout the period of observation or until the death of the animal. In the spleen, however, after the maximum is reached within two weeks or so, the number of bacilli slowly decreases in all cases, even when the infection eventually leads to a fatal outcome.

Kim (2) also observed the fate of tubercle bacilli quantitatively in these organs as well as in the kidney and liver. He concluded that the results in the lung and spleen were in accordance with the findings of Pierce and associates and that, in the kidney and liver, the fate of the bacilli was similar to that seen in the lung and the spleen, respectively.

The present writer has been interested in specific organ-parasite relationships and has pursued the interaction between the host cell and the parasite by challenging the lung tissue cultured *in vitro* with two virulent strains of tubercle bacilli.

Materials and Methods

Embryonic lung peripheries of guinea pigs, pregnant forty to fifty days, were used. Some tissues were used without the pleural covering. When cultured *in vitro*, the tissues were chiefly composed of three kinds of cells: epithelial cells, macrophages, and spindle cells. As stated in the preceding paper (3), the writer considered most of the epithelial cells on the culture surface to be of alveolar epithelial origin, and the macrophages to be the alveolar phagocytes called "dust cells" and "septal cells." The spindle cells must originate from capillary endothelial cells and fibroblasts. The method of tissue culture in this experiment is described in detail in the foregoing paper (3) and needs no further comment.

Mycobacterium tuberculosis var. *hominis*, strain Kurono, is fully virulent; more virulent for mice and guinea pigs than our laboratory stock culture of H37Rv. Ravenel is a strain received from the National Institute of Health of Japan.

The suspensions for infection were prepared from a ten-day culture on Ogawa's solid egg medium. The incubated bacilli were placed in a 50-ml. flask for grinding by glass beads 5 to 6 mm. in diameter. They were then placed in Tyrode's solution for two five-minute centrifugations of 1,200 r.p.m. The resulting supernatant contained chiefly individually separated cells. The suspension thus treated was diluted ten times with the nutrient medium. This diluted suspension contained about 10^6 viable units.

The tissue, cultured forty-eight to seventy-two hours at 37°C., was brought into contact with this suspension for four days. The culture was then transferred into the bacterium-free nutrient; and this procedure was repeated every third day thereafter. The pH of the medium was adjusted to 7.4–7.6.

Every twenty-four hours following the first introduction of bacilli, two samples from each strain were successively examined. They were fixed in 10 per cent neutral formalin and stained by the Ziehl-Neelsen technique.

Results

The most aggressive reaction *in vitro* against fully virulent tubercle bacilli was shown by macrophages. They ingested many bacilli in twenty-four hours and were found among spindle cells. The ingested bacilli in their earlier stage of infection were sometimes granular in appearance (figure 1) but, by about the third day of infection, the cells seemed to be affected by the virulent tubercle bacilli. They were then almost necrotic (figure 2). In about five days almost all macrophages, whether ingesting many or few bacilli, disappeared from the culture surface. The bacilli that destroyed cells were seen extracellularly, when extracellular cord formation with multiplication rapidly took place (figure 3).

On the contrary, the epithelial cells were not so active against the bacilli as in the case of macrophages, and phagocytosis was limited. A sample of epithelial cells two days after infection showed one to several bacilli dispersed in the protoplasm,

[1] From Kitasato Institute for Infectious Diseases, Tokyo, Japan.

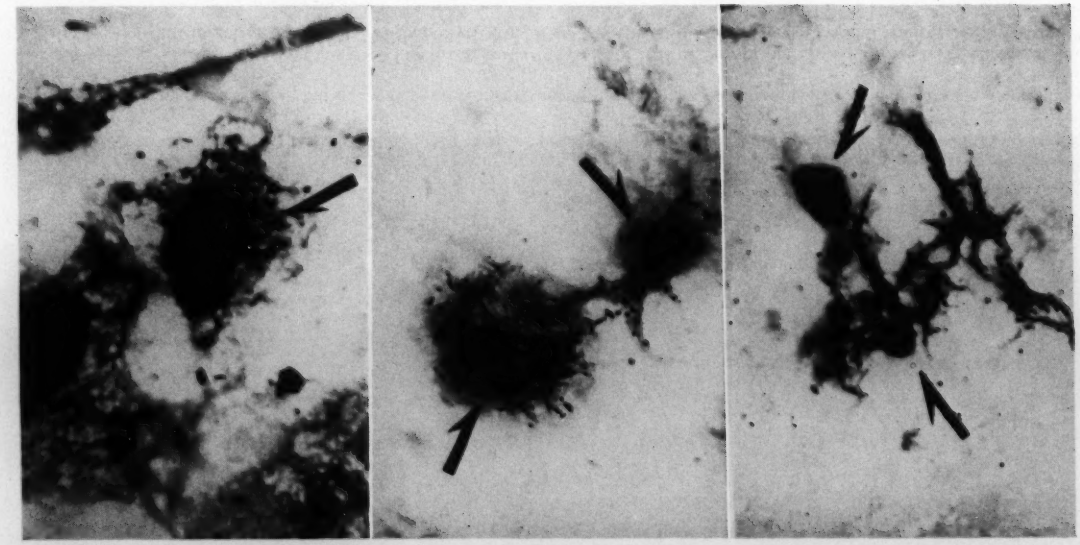

FIG. 1 (Left). Kurono, forty-eight-hour infection. (1500×) Bacilli (↑) ingested by macrophages are usually granular.

FIG. 2 (Center). Kurono, seventy-two-hour infection. (1500×) Macrophages (↑) that have ingested bacilli are affected by the virulent tubercle bacilli and degenerate. But the bacilli gradually become rodlike.

FIG. 3 (Right). Kurono, five-day infection. (1500×) Ingesting bacilli, macrophages at this stage have almost completely been destroyed. A bit of nuclear remnant (↑) is seen. The bacilli are now multiplying to form an extracellular cord.

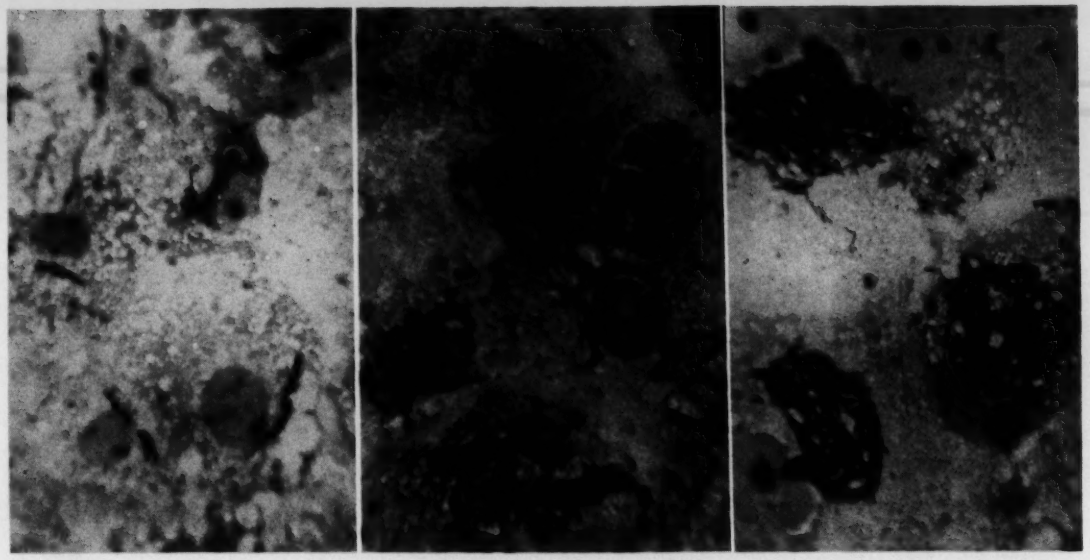

FIG. 4 (Left). Kurono, four-day infection. (1200×) Bacilli in the cell body of the epithelium have the tendency to form a cord near the nucleus. The cord is still short.

FIG. 5 (Center). Kurono, seven-day infection. (1200×) The intracellular bacterial cords in the epithelium are shown to be elongated and enlarged.

FIG. 6 (Right). Kurono, ten-day infection. (1200×) The bacterial growth in the epithelium is remarkable and the intracellular cords appear more complicated.

but it was difficult to tell whether the bacilli were really intracellular or extracellular. The sample of a four- to five-day culture, however, demonstrated bacilli proliferating in the characteristic cords (figure 4). These findings in the epithelial cells were rather rare at this stage. The bacilli were likely to form the cords in the immediate vicinity of the nucleus, and the cords sometimes looped around the nucleus. This proved that a few bacilli ingested by the cells at the initial stage of infection had proliferated intracellularly.

Seven days after infection the bacilli in the cells showed many enlarged and elongated serpentine cords (figure 5). It was also observed that the number of cells containing the bacilli was also increased; in certain areas, to 70 to 80 per cent of the epithelial cells contained the bacterial cords.

The picture on the tenth day of infection showed more complicated cords than that on the seventh day (figure 6), but the intracellular cords were distinctly different from extracellular cords. This difference is shown in figure 7. Intracellular cords were somewhat inhibited and not as free as extracellular cords.

Often the bacterial growth was so conspicuous as to fill up the entire body of the cell so that the nucleus was pushed aside, causing pycnosis. No remarkable degeneration of the cells, however, could be seen even in this stage of infection except of those cells which were filled with the bacilli or whose nuclei were surrounded by the cords.

In some spindle cells, intracellular bacilli were also clearly found about the fifth day of infection, but there was almost no such typical formation of cords as seen in the epithelial cells. They were usually on one side or both sides of the nucleus and sometimes filled the entire cell with the

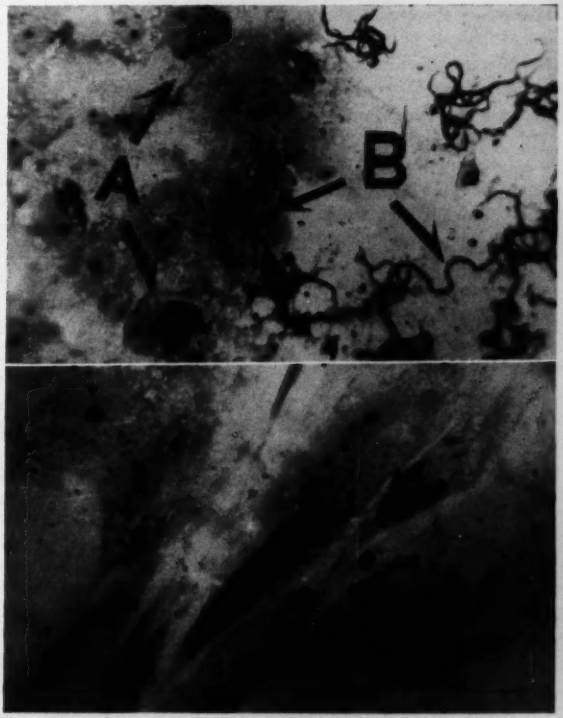

FIG. 7 (Upper). Kurono, ten-day infection. (800×) Intra-epithelial cell cords (*A*) are clearly differentiated from extracellular cords (*B*). While *B* is serpentine, *A* is rather glomerular.

FIG. 8 (Lower). Kurono, seven-day infection. (1200×) Spindle cells also offer a fairly good ground for bacterial growth. No cord formation is seen in the epithelium, but the bacilli are seen parallel along their long axes.

bacterial clump (figure 8). Occasionally bacilli filled the cell body on the seventh day of infection. However, as the cells were not as phagocytic as macrophages against tubercle bacilli, the intracellular multiplication without forming cords must have caused this kind of finding. Spindle cells kept bacilli in the body for about a week without being affected by the bacilli morphologically, but about half of them were destroyed by the tenth day after infection.

Although the findings above were obtained in the experiment with the Kurono strain, the Ravenel strain gave practically similar results.

Discussion

As observed by many investigators (4–6), macrophages are markedly phagocytic against tubercle bacilli. The bacilli occasionally appeared to be granular in these cells. The ingested bacilli never formed serpentine cords in these cells under the normal conditions. Maximow (7) reported that bovine virulent tubercle bacilli formed filaments in the histiocytes *in vitro*. However, the present writer cannot agree with him completely that the filaments are true intracellular cords.

In contrast with these virulent strains, in an experiment with attenuated strains such as R1Rv, BCG, macrophages survived longer with many bacilli in their cell bodies (8). In this respect, Dubos (9), quoting Bloch's experiment (10), suggested that virulence is correlated with the ability of the bacilli to cause injury to the phagocytic cells. Furthermore, reports made by Suter (11) and Mackaness and associates (12) on early degeneration and destruction of mononuclear phagocytes by virulent strains confirmed the present writer in his assumption that macrophages in general would be seriously injured by virulent tubercle bacilli.

According to Low and Sampaio (13), alveolar epithelial cells totally lack phagocytizing ability. In the present experiment it was shown that the alveolar epithelial cells ingested tubercle bacilli *in vitro*, although their phagocytizing ability was not so positive as that of macrophages. In the epithelial cell, contrary to the macrophage, the bacilli vigorously multiplied forming intracellular cords near or around the nuclei just as in the case of omentum cells observed by Woodruff (14), and of HeLa cells reported by Shepard (15). This fact would suggest that the epithelial cells provide a favorable environment for the growth of virulent tubercle bacilli. The epithelial cells, however, did not noticeably degenerate for a long span of time, while fully virulent tubercle bacilli were multiplying with a high growth rate. Spindle cells occasionally ingest tubercle bacilli, but the bacilli in them do not form typical cords; they usually appear as bacterial clumps.

Referring more specifically to epithelial cells and macrophages, it is said that monocytes have a lipid-digesting enzyme similar to lipase (16). Such an enzyme has not been reported in epithelial cells. Inasmuch as macrophages and monocytes are the cells of one category, macrophages would have the same enzyme. If so, it seems natural that even the virulent tubercle bacilli could not form intracellular cords, because this enzyme would digest the lipid fraction of the bacilli, which surrounds the surface of virulent tubercle bacilli and is related to cord formation (17). This would, in turn, cause early degeneration of the cells because the toxic components of the bacilli (17–19) would be liberated by the bacteriolytic action of these cells initiated by the lipolytic function of the enzyme.

There is a difference in the epithelial cells. The bacilli seem to stay in the cell as harmless foreign bodies, which would allow both the cells and the bacilli to coexist for a considerable period with little friction between the two.

Whatever the cause of these differences of host-parasite relationship may be, the fact is worthy of attention that the epithelial cell sheets, which offer comparatively favorable environment to the bacilli, cover the pulmonary alveolar wall. This would carry weight in explaining the mechanism of primary infection of pulmonary tuberculosis resulting from a very few bacilli that have reached the lung through the air passage. Frequent bronchogenic spread in pulmonary tuberculosis might also be similarly explained.

Summary

Three types of cells, macrophages, epithelial cells, and spindle cells, grown in cultures of embryonic lung tissue of the guinea pig, were subjected to contact with virulent strains of tubercle bacilli, and the cell-bacilli relations were studied.

The macrophage had the strongest phagocytosis of the three. This cell also showed bacilli-disintegrating action in the early stage of infection, but it was markedly injured by the intracellular bacilli.

In contrast to the macrophage, the epithelial cell could ingest only few bacilli; but these few intracellular bacilli could multiply at a high growth rate, forming characteristic cords. Despite this vigorous growth of virulent bacilli, the cytopathic effect of the bacilli was least on the epithelial cell.

The spindle cell, also, could sometimes ingest a few bacilli, but the bacilli could not form intracellular cords.

Sumario

Estudios de la Interacción entre las Células y los Parásitos del Huésped

Tres formas de glóbulos—macrófagos, epiteliocitos y células fusiformes—criadas en cultivos de tejido pulmonar embrionario de cobayo, fueron expuestas al contacto con cepas virulentas de bacilos tuberculosos, para estudiar las relaciones entre glóbulos y bacilos.

De los tres, el macrófago desplegó la fagocitosis más poderosa. Este glóbulo también reveló acción desintegrante de los bacilos en la fase incipiente de la infección, pero fué averiado intensamente por los bacilos intracelulares.

En contraposición al macrófago, el epiteliocito no pudo ingerir más que pocos bacilos; pero estos pocos bacilos intracelulares se podían multiplicar con mucha rapidez, formando típicos cordones. A pesar de esa enérgica proliferación de bacilos virulentos, el efecto citopático alcanzó su mínimo en los epiteliocitos.

La célula fusiforme pudo también, algunas veces, ingerir unos pocos bacilos, pero éstos no podían formar cordones intracelulares.

Resume

Etudes des réactions réciproques de la cellule hôte et du parasite

Trois types de cellules—macrophages, cellules épithéliales et fibroblastes—dont la croissance a eu lieu dans des cultures de tissu pulmonaire de l'embryon de cobaye, ont été mises au contact de souches virulentes de bacilles tuberculeux et les rapports cellules-bacilles ont été étudiés.

Des trois, les macrophages avaient le pouvoir phagocytaire le plus grand. Cette cellule a démontré aussi une action désingégrante vis-à-vis des bacilles au premier stade de l'infection, mais elle était nettement endommagée par les bacilles intracellulaires.

La cellule épithéliale, par contre, ne pouvait ingérer qu'un petit nombre de bacilles, mais ces quelques bacilles intracellulaires pouvaient se multiplier à un rythme accéléré, formant des cordons caractéristiques. Malgré ce développement énergique de bacilles virulents, l'effect cytopathique des bacilles était le moins marqué sur la cellule épithéliale.

Les fibroblastes pouvaient, eux aussi, parfois ingérer quelques bacilles, mais ceux-ci ne pouvaient pas former de cordons intracellulaires.

Acknowledgment

The writer gratefully acknowledges his appreciation to Dr. E. S. Han whose encouragement made this paper possible.

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FURTHER STUDIES ON AUTO-TISSUE SUBSTANCES IN TUBERCULOUS RABBITS[1, 2]

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(Received for publication May 18, 1959)

INTRODUCTION

From previous work in this laboratory (1) it was learned that lung-specific antibodies could be produced in rabbits by intraperitoneal injections of homologous tuberculous lung suspensions or by normal lung and killed tubercle bacilli in Freund's adjuvant. In addition, antibodies with apparent specificities for other organs were demonstrated in the same serum samples. Injection of normal lung alone did not stimulate the production of these anti-tissue substances.

The purpose of the present report is to describe the results of examining serums of animals infected with tubercle bacilli for lung-specific antibodies and to determine what effect, if any, these substances may have in tuberculous disease.

EXPERIMENTAL

Occurrence of anti-lung substances in tuberculous disease: The question arose whether lung-specific antibodies are produced in rabbits infected with virulent tubercle bacilli alone.

Four pairs of rabbits received graded intravenous inocula of viable *Mycobacterium tuberculosis* (var. Ravenel) in 0.25, 0.5, 1.0, and 2.0 mg. doses, respectively. Weekly serum samples were examined for lung antibodies by the complement-fixation method, using a previously described ultrasonic irradiated normal lung antigen.

In table 1 it may be seen that the critical dose of tubercle bacilli necessary for the serum demonstration of these antibodies is between 0.5 and 1.0 mg. The titers in all instances were very low and demonstrable for only a short time. These same serums were also examined for the presence of kidney-specific antibodies with ultrasound-treated kidney antigen, but these antibodies were detected in only minimal quantities in the higher-titered lung antibody serums from rabbits Nos. 908 and 909. In a previous paper concerning these matters (1), it was reported that the demonstration of these substances in diseased animals was unsuccessful. This statement was based upon the observations of animals injected with 0.1 mg. of virulent tubercle bacilli which apparently is below the critical dose, at least under the conditions employed for measurement.

Effect of lung antibodies in the normal rabbit: Since the possible role of these tissue-specific antibodies in tuberculous infection was not obvious, the following series of experiments were conducted to establish their significance.

Experiment 1: Each of two pairs of rabbits was injected intravenously with 5.0 ml. of either high-titered anti-lung serum or with normal serum. Immediately following the injections, the rabbits which had received the immune serums demonstrated rapid shallow breathing and excessive urination.

Forty-eight hours after injection, the animals were sacrificed and representative tissues were examined histologically.

In general, the lungs from the rabbits which had received anti-lung serum showed an accumulation of reactions in the peribronchial regions, some mild interstitial reactions, an increase in mononuclear cells, and some evidence of pulmonary edema. The kidneys were swollen and exhibited some evidence of tubular degeneration and hemorrhage. Tissues from the rabbits which had received normal serum exhibited less marked reactions. It was thought that iso-antibodies might have contributed to some of the pathology.

Lungs from a normal rabbit injected with rat anti-lung serum, prepared for purposes of control, revealed areas of slight congestion and areas suggestive of bronchopenumonia. Similar results were obtained by Frick (2), who injected rats with a lung tissue-Freund's type of adjuvant emulsion. Pneumonic lesions due to antigen-antibody reactions have also been obtained by Schlecht and Schwentker (3).

Experiment 2: Six rabbits were employed. Three of these rabbits received 5.0-ml. intravenous in-

[1] From the Department of Bacteriology, The Ohio State University, Columbus 10, Ohio.

[2] This study was supported in part by a grant from the Ohio Tuberculosis and Health Association.

TABLE 1

EFFECT OF DOSAGE OF MYCOBACTERIA ON THE PRODUCTION OF ANTI-LUNG SUBSTANCES

abbit umber	Dose	Complement-Fixation Titers (weeks)						
		BL	1	2	3	4	5	6
	mg.							
640	0.25	—	—	—	—	—	...	...
641	0.25	—	—	—	—	—	—	—
905	0.5	—	—	8	8	—	—	—
907	0.5	—	—	—	8	—	—	—
908	1.0	—	—	8*	4	—	—	—
909	1.0	—	—	4	8*	2	2	16
913	2.0	—	—	4	—	—	—	—
917	2.0	—	—	16	—	—	—	—

BL refers to preinjection bleeding.
... Animal died.
* Serum also contained kidney-specific antibodies (1:2).

jections of pooled high-titered anti-lung serums and 3 received pooled normal serums. Forty-eight hours later each rabbit was injected with 0.01 mg. of virulent tubercle bacilli by the same route. The arbitrary dose was selected because it was believed that this amount would be enough to cause a minimal infection and thus enable the effects of the serums to be more easily realized. The animals were sacrificed after two weeks, and representative tissues were examined grossly as well as histologically.

Grossly, the lungs from the rabbits which had received normal serum before infection were typical of those from tuberculous rabbits. A few discrete tubercles were readily discernible, but no other significant changes were observed. However, the lungs from the rabbits which had received immune serum prior to infection demonstrated marked hemorrhagic and inflammatory qualities, along with a miliary type of disease as evidenced by numerous pinpoint tubercles. Regardless of the prior treatment rendered the rabbits, no distinctive tissue change was found in the kidney specimens.

In the lung sections, certain tissue changes were of interest. For example, lungs from an animal previously injected with immune serum prior to becoming infected showed an occasional miliary type of nodule, scattered areas of hemorrhage, and some inflammatory involvement of the bronchioles. A lung from a rabbit similarly treated with normal serum exhibited tissue changes more in keeping with classical tuberculosis (figure 1).

It is not to be inferred that the reactions just described were characteristic of the tissues from all animals which had received the same treatment. It must be emphasized that generalizations were made concerning these specimens, since reactions were not always "textbook-defined and -differentiated." Whether or not this is due to selecting an inopportune time for sacrifice or to an error in experimental design requires further investigation.

Experiment 3: To eliminate from consideration the possible effect of iso-antibodies from the donor serum, each serum aliquot in the following experment was adsorbed with erythrocytes from the intended recipient normal rabbit before intravenous administration. Four milliliters of properly adsorbed serum of the following types were injected into the respective normal animals: pooled normal serum (No. 822); pooled immune serums (Nos. 807 and 802); pooled immune serum previously adsorbed with dried tubercle bacilli (No. 821); pooled immune serum previously adsorbed with normal kidney sediment (No. 811); pooled immune serum previously adsorbed with normal lung sediment (No. 823); and pooled immune serum previously adsorbed with dried tubercle bacilli and normal lung and kidney sediments (No. 817).

Forty-eight hours after injection of these serums, all rabbits except No. 807 received an intravenous injection of 0.01 mg. of virulent tubercle bacilli. After two weeks the animals were sacrificed and the tissues examined as in the previous experiments.

As before, the tissues from the animals which had received serums which contained anti-lung substances, i.e., those not adsorbed prior to infection, revealed grossly hemorrhagic qualities and numerous pinpoint tubercles suggesting a miliary type of infection. Tissues from the animals previously treated with normal serums, on the other hand, were characterized by more classical tuberculous pathology.

After gross examination the respective lungs were fixed in 10 per cent formalin, embedded in paraffin, and sectioned. Companion sections from each tissue specimen were placed on each of two slides. One section was stained by the eosin-hematoxylin method and the other by an acid-fast staining technique. In this way the tissue sections could be compared with the quantities of mycobacteria in each lesion.

Although various tissues were examined, the tissue selected for the principal study was the lung. Again, generalizations of the over-all reactions must be made.

When the iso-antibodies were eliminated by proper adsorptions, the histopathologic effects of the immune serum pretreatment became more evident. In contrast to the tissues of the animals pretreated with normal serum (*see* figure 1) or with serum from which all lung-specific antibodies had been removed, the immune serum pretreated tissues were characterized by tissue sections associated with the miliary type of disease (figure 2). It is of interest that, when the tuberculous antibodies were removed from the pooled immune serums by adsorption with killed tubercle bacilli and these serums were employed for the pretreatment, the tissue presented a pathologic

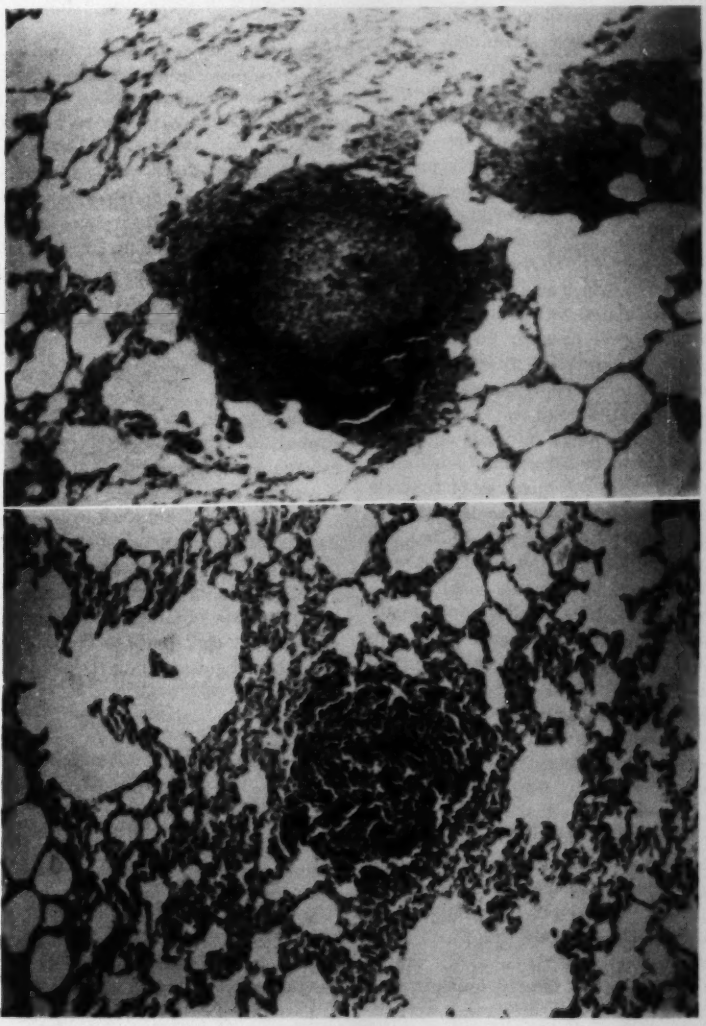

FIG. 1. Typical tuberculous lesion from rabbit which received pooled normal serum prior to infection.
FIG. 2. Acute lesion from rabbit which received immune serum prior to infection. Demonstrates a poorer tissue response.

picture similar to those that received whole immune serum except that the microorganisms were more abundant in the lesions.

Discussion

Since it has been shown that antibodies specific for normal lung tissue can be produced artificially in conjunction with tuberculous processes and/or components, it seemed necessary to establish the possible existence of these substances under normal conditions. The results have indicated that demonstration of lung-specific antibodies is possible in the serums of animals that have been infected solely with virulent tubercle bacilli. Since these antibodies were transiently present and then only in low titers, it may be inferred that they became localized soon after their production.

Although there seems to be a minimal dose of tubercle bacilli below which no detectable serum antibodies are stimulated, it is of interest that these substances appear as early as two weeks after inoculation providing the infective dose of organisms is adequate. It seems that the production of lung-specific antibodies may be definitely associated with tuberculous infection or, more precisely, the serum demonstration of such antibodies is dependent on the quantity of tubercle bacilli comprising the infective dose or the quantity attained during active infection. Indeed, the serum from a late bleeding from one rabbit with extremely advanced tuberculosis exhibited a sharp rise in anti-lung titer shortly before death.

The localization of these anti-lung substances was studied indirectly by challenging normal rabbits with variously treated immune serums and examining their tissues for pathologic changes after a suitable incubation period. The classical Masugi nephritis (4) is demonstrated very readily by this method.

It should be re-emphasized that the histologic examination of the test animals was made principally of lung tissue and was directed toward determining the extent of granuloma formation with a rough approximation of the relative numbers of organisms present in the lesions.

Differences in the organization of the granulomas from the variously treated animals were noted. Including among these differences were the extent of epithelioid reaction, necrosis, and polymorphonuclear neutrophilic infiltration as well as the number of tubercle bacilli discernible in the lesions. Many of the animals shared these pathologic findings. Depending on the specific treatment rendered the particular serum introduced prior to challenge, however, certain of the pathologic aspects were emphasized. Since rabbits demonstrate such a marked range of reaction to insult, it is obvious that larger groups of animals need to be studied in order to define more clearly the influence of tissue antibodies on subsequent tuberculous infection.

From the data recounted here, it is obvious that it is possible to produce auto-tissue antibodies in experimental tuberculosis. A principal deterrent to establishing a possible existence or role of similar antibodies in the naturally occurring human disease is a lack of definitive pathologic criteria, although it is reasonable to assume that such a mechanism may exist. However, more conclusive evidence awaits further experimentation and the establishment of criteria for the pathology which, of necessity, may depart from the classic concepts now accepted.

Summary

Lung-specific antibodies have been demonstrated in rabbits infected with experimental tuberculosis. Apparently, a minimal dose is necessary for their production.

Histopathologic studies were attempted on tissues from rabbits that had been injected with normal or anti-lung serums prior to a minimal infective dose of virulent tubercle bacilli. Some indication was obtained that these anti-lung substances may contribute to the infectious process. More investigation along these lines is indicated.

Sumario

Nuevos Estudios sobre Substancias de Auto-Tejidos en los Conejos Tuberculosos

En los conejos infectados con tuberculosis experimental se han encontrado anticuerpos específicos para el pulmón. Al parecer, se necesita una dosis mínima para producirlos.

Se iniciaron estudios histopatológicos en tejidos procedentes de conejos a los que se habían inyectado sueros normales o anti-pulmón antes de una dosis infecciosa mínima de bacilos tuberculosos virulentos. Obtúvose alguna indicación de que estas substancias anti-pulmón pueden contribuir al proceso infeccioso. Hállase indicada más investigación en ese sentido.

Resume

Etudes complementaires sur les substances produites par le poumon lui-meme chez des lapins tuberculeux

Il a été demontré chez des lapins présentant une tuberculose expérimentale que le poumon produisait ses propres anticorps. Apparemment, une dose minimum est necessaire pour leur production. On a tenté des études histologiques des tissus de lapins qui ont reçu une injection de sérum ordinaire ou de sérum contenant ces anticorps, avant de recevoir une injection de bacilles tuberculeux virulents. D' aprés ceci, ces substances: Anticorps "anti-poumon" pourraient avoir un role dans le processus infectieux. Des recherches ultérieures sont encore désirables dans ce domaine.

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RAPID IDENTIFICATION OF MYCOBACTERIAL COLONIES[1]

I. The Auramine Test

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(Received for publication June 8, 1959)

Introduction

The staining of tubercle bacilli with the dye auramine was demonstrated by Hagemann (1) in 1938. Richards and Miller (2) in 1941 showed that auramine could be used for staining acid-fast bacilli present on smears with the use of fluorescence microscopy technique.

The application of fluorescence microscopy for the detection of tubercle bacilli was further investigated by Richards, Kline, and Leach (3), Bogen (4), Thompson (5), and Lind and Shaughnessy (6). The conclusion reached by these workers indicated that auramine was a specific dye for acid-fast organisms in the same manner as was the basic fuchsin (Ziehl-Neelsen) stain. There was also general agreement that the auramine fluorescence technique was superior to the Ziehl-Neelsen method in comparative studies. Therefore, if individual acid-fast organisms could be stained with auramine, it appeared reasonable that intact acid-fast colonies could likewise be stained.

For some time, it had been the goal of this laboratory to stain colonies *in situ* on the surface of a culture medium. A translucent oleic–acid–albumin agar type of medium used in this laboratory appeared to be ideally suited for this new staining technique, at the same time allowing *in situ* observation of stained colonies for morphologic characteristics and cording.

It was observed in preliminary studies with auramine that a solution of this dye became nearly colorless upon the addition of an alkali. Because auramine solutions are brilliant yellow at acid or neutral pH, the advantage of this color contrast was further investigated, and formed the basis for the staining technique by the auramine method.

It was speculated that the application of the auramine test technique could perhaps result in the differentiation of some categories of acid-fast organisms, and with this possibility in mind the present study was undertaken.

[1] From Arroyo Del Valle Sanatorium, Livermore, California.

Materials and Methods

In preliminary tests a total of 59 strains of Mycobacterium cultures were studied. Most of these were kindly supplied by Dr. Lawrence Wayne,[2] Dr. Ernest Runyon,[3] and Dr. Seymour Froman.[4] Forty-three clinical strains of *M. tuberculosis* isolated by this laboratory from previously diagnosed patients were also included in this study.

Three methods were used in testing all strains.

Method I: In this series of experiments a solution was prepared by dissolving 0.1 gm of auramine O.C.I. No. 655[5] in 100 ml. of 95 per cent ethyl alcohol and filtering. Half-inch discs specially prepared from Whatman No. 1 filter chromatography paper were impregnated with this solution and carefully dried. Similar control discs were also specially prepared without auramine. The discs were then placed in horizontal rows in sets of three: an auramine-treated test disc; a control test disc without auramine; and an auramine-treated control disc. The container used for the discs consisted of a transparent series of plastic wells. Any desired combination of these plastic wells could be obtained by cutting a section from a special Disposo-Tray Model 96-WS.[6]

Submitted cultures were then tested as follows: A small quantity of culture growth was removed with the end portion of a sterile wooden applicator. The specimen was then worked into the fibers of the auramine-treated disc by firmly pressing and twirling the applicator stick until only a shadow of the material remained on the surface of the disc. This procedure was repeated on the control disc without auramine, using another applicator stick. The auramine-treated control disc remained intact. Two drops (approximately 0.06 ml.) of a 0.5 N aqueous sodium hydroxide solution were then allowed to drop on each disc from a medicine dropper, and the test was read after two minutes while the disc was still moist.

An auramine-positive test exhibited a bright yellow area which persisted after two minutes against a white background of the disc. Discs

[2] Veterans Administration Hospital, Salt Lake City, Utah.

[3] Veterans Administration Hospital, San Fernando, California.

[4] Olive View Sanatorium, Olive View, California.

[5] Manufactured by the National Aniline Division, New York, New York.

[6] Manufactured by Linbro Chemical Co., Inc., New Haven, Connecticut.

showing weak positive macroscopic reactions could be readily examined in their transparent wells microscopically, using a 50 times magnification with a blue glass filter.

A test was interpreted as auramine negative if the bright yellow color did not develop when viewed macroscopically. Microscopically, the material on the disc appeared grayish-green or gray. Discs showing a yellow area which decolorized within two minutes were also grouped in the negative category. Method I will hereafter be referred to as the auramine disc test.

Method II: In these series of tests the same filtered alcoholic solution of auramine was employed (*see* Method I).

All submitted strains were subcultured on a translucent oleic-acid–albumin agar type of medium prepared in this laboratory. Cultures were incubated at 37° C. in sealed 60-mm. Petri dishes. When sufficient growth had developed on each plate to allow the detection of colony morphology microscopically, the plates were subjected to the following test:

Equal volumes of alcoholic auramine solution and 1.0 N aqueous sodium hydroxide were mixed just prior to the test, resulting in a clear slightly tinted reagent. Two to three milliliters of this reagent were then allowed to flood the open Petri dish. Within one minute the auramine-positive colonies selectively began to extract the dye from the alkaline reagent and stained a brilliant yellow *in situ*, contrasting against an almost colorless background of the culture medium.

This test was interpreted as auramine positive if the yellow stain persisted after one minute within the colonies. The increasing depth of color in the colonies could be observed microscopically immediately upon the addition of the reagent. This staining technique appeared to impart better definition to colony morphology and cording. When cord formations could not be definitely distinguished with low-power magnification, the reagent was decanted and a 22 by 22 mm. plastic coverslip was placed on the surface of the medium, permitting the colonies to be examined at higher magnifications.

By this method, auramine-negative colonies did not visibly extract the auramine dye from the reagent in the presence of an alkali and alcohol. Colonies staining yellow but decolorizing within one minute were also designated as auramine negative. Method II hereafter will be referred to as the auramine flood test.

Method III: After the auramine disc test was completed, all discs used in that test were left intact in their respective plastic wells and incubated at 37° C. for approximately two hours.

By a simple fluorescent technique using a very inexpensive ultraviolet blacklight lamp,[7] the various strains of mycobacteria on the discs exhibited various types of fluorescence against a nonfluorescing lavender background when observed in the dark. Method III hereafter will be referred to as the auramine fluorescence test.

The 43 clinical strains of *M. tuberculosis* used in this study were isolated under routine laboratory conditions from the sputum of 100 different patients. The standard sodium hydroxide method was used for concentrating the specimens. All concentrates were routinely cultured in 60-mm. Petri dishes on an oleic-acid–albumin agar type of medium and incubated at 37° C. for six weeks. After the incubation period all cultures were routinely exposed to light to rule out photochromogens. The 100 plates were then submitted to a thorough routine microscopic examination for mycobacterial colonies and cording. Positive plates were tested by the three previously described methods, while all presumably negative and contaminated cultures were tested by the auramine flood test.

A group of nonacid-fast organisms frequently encountered in clinical laboratories was also included for comparison in this series of experiments and was tested by Methods I, II, and III.

Results

The various strains of organisms employed in these experiments were tested by three auramine methods and the results were compared with the microcolonial test of Wayne (7, 8). These results are presented in tabulated form (tables 1 to 7), and the breakdown of the various groups of organisms tested were as follows:

I. Nonacid-fast group
II. Standard strains of mycobacteria group
III. Photochromogen group
IV. Nonphotochromogen group
V. Skotochromogen group
VI. Miscellaneous acid-fast group
VII. *M. tuberculosis* group

I. Nonacid-fast group: The results in table 1 indicated that nonacid-fast organisms were negative by all three auramine methods. Various fungi

TABLE 1

Reactions of Nonacid-fast Organisms with Auramine Methods

Organism	Source	Auramine Disc Test	Auramine Flood Test	Auramine Fluorescence Test
S. albus	All clinical laboratory isolates	—	—	NF
S. aureus		—	—	NF
S. citreus		—	—	NF
E. coli		—	—	NF
B. pyocyaneus		—	—	NF
Monilia		—	—	NF
Various fungi		—	—	NF

NF = No visible fluorescence

[7] Manufactured by The Stroblite Co., New York, New York.

TABLE 2

REACTIONS OF STANDARD STRAINS OF MYCOBACTERIA WITH AURAMINE METHODS

Strain	Source	Microcolonial Test		Auramine Disc Test	Auramine Flood Test	Cord on 0-A-AA Media	Auramine Fluorescence Test
		Cord	Neutral Red				
M. tuberculosis, H37Rv	R	+	+	+	+	+	G
M. tuberculosis, H37Ra	R	−	+	+	+	−	G
M. tuberculosis, Rlrv	R	−	+	+	+	−	G
M. tuberculosis (Ravenel)	T	+	+	+	+	+	G
M. avium, Kirschberg	R	−	+*	+	+*	−	G
BCG #793	P	+	+	+	+	+	G

* Weak positive reactions

G—greenish; O-A-AA—oleic-acid-albumin agar; R—Runyon (Veterans Administration Hospital), Utah; T—Trudeau Laboratory, New York; P—Phipps Institute, Pennsylvania.

TABLE 3

REACTIONS OF PHOTOCHROMOGENS WITH AURAMINE METHODS

Photochromogen Number	Source	Microcolonial Test		Auramine Disc Test	Auramine Flood Test	Cord on O-A-AA Media	Auramine Fluorescence Test
		Cord	Neutral Red				
1	R	+	+	+	+	+	G
4	R	+	+	+	+	+	G
8	R	+	+	+	+	+	YO
16	R	+	+	+	+	+	YO
18	R	+	+	+	+	+	G
21	R	+	+	+	+	+	YO
22	R	+	+	+	+	+	G
24	R	+	+	+	+	+	G
26	R	+	+	+	+	+	YO
47	W	+	+	+	±	+	G
50	W	+	+	+	+	+	YO
55P	W	+	+	+	+	+	YO
96	W	+	+	+	+	+	YO
159	W	+	+	+	+	+	G

±—doubtful reaction

G—greenish

YO—yellow to orange

R—Runyon (Veterans Administration Hospital), Utah

W—Wayne, (Veterans Administration Hospital), California.

appearing as contaminants on the culture medium have failed thus far to yield positive auramine tests. Monilia was the most frequent contaminant encountered in routine laboratory culturing methods for *M. tuberculosis*.

II. Standard strains of mycobacteria group: A close parallel was observed between the auramine (disc and flood tests) and the neutral red reaction in the 6 standard strains tested. Cord formation also appeared to correspond to the microcolonial method. Cording was determined microscopically on oleic-acid-albumin agar medium, using high-power magnification. Greenish fluorescence was observed with all six strains in this group.

III. Photochromogen group: A group of 14

TABLE 4

REACTIONS OF NONPHOTOCHROMOGENS WITH AURAMINE METHODS

Nonphotochromogen Number	Source	Microcolonial Test		Auramine Disc Test	Auramine Flood Test	Cord on 0-A-AA Media	Auramine Fluorescence Test
		Cord	Neutral Red				
2	R	−	+	+	+	−	YO
3	R	−	+	+	+	−	G
7	R	−	+	+	+	−	G
17	R	−	+*	+*	+	−	G
20	R	−	+	+	+	−	YO
23	R	−	+*	+*	+	−	G
25	R	−	+*	+	+	−	G
53	W	−	+	+	+	−	G
67	W	−	+	+	+	−	G
69	W	−	+	+	+	−	G
163	W	−	+	+	+	−	G
169	W	−	+	+	+	−	YO
173	W	−	+	+	±	−	YO
174	W	−	+	+	+	−	YO
247	W	−	+	+*	±	−	G

* Weak positive reactions.

± Doubtful reactions.

G—greenish.

YO—yellow to orange.

R—Runyon (Veterans Administration Hospital), Utah.

W—Wayne (Veterans Administration Hospital), California.

TABLE 5

REACTIONS OF SKOTOCHROMOGENS WITH AURAMINE METHODS

Skotochromogen Number	Source	Microcolonial Test		Auramine Disc Test	Auramine Flood Test	Cord on 0-A-AA Media	Auramine Fluorescence Test
		Cord	Neutral Red				
5	R	−	+	+	±	−	G
6	R	−	+	+	+	−	FNF
15	R	−	+	+	−	−	FNF
19	R	−	+	+	+	−	G
189	W	−	+	+	+	−	G
190	W	−	+	+	±	−	FNF
191	W	−	+	+	+	−	FNF
194	W	−	+	+	+	−	G
222	W	−	+	+	+	−	G
269	W	−	+	+	+	−	G

± Doubtful reactions.
G—greenish.
FNF—faint to no fluorescence
R—Runyon (Veterans Administration Hospital), Utah.
W—Wayne (Veterans Administration Hospital), California.

photochromogens showed excellent correlation of results between the auramine (disc and flood tests) and the neutral red reaction. Number 47 was the only strain that presented a doubtful reaction by the auramine flood test. All photochromogens tested appeared to be corded. Fluorescence in this group was evenly divided between the greenish and the yellow to orange types.

IV. Nonphotochromogen group: Fifteen strains tested in this group were generally in good agreement when results by the auramine disc and flood tests were compared with the neutral red reaction. However, two discrepancies were observed with strains Nos. 173 and 247. These produced doubtful reactions by the flood technique. It may be of interest to note that there was excellent correlation of results between the auramine disc test and the neutral red reaction even to the point of weakly positive reactions. All nonphotochromogens tested appeared to be noncorded.

In this group, greenish fluorescence was ob-

TABLE 6

REACTIONS OF MISCELLANEOUS ACID-FAST STRAINS WITH AURAMINE METHODS

Identification Number	Type of Organism	Source	Microcolonial Test		Auramine Disc Test	Auramine Flood Test	Cord on 0-A-AA Media	Auramine Fluorescence Test
			Cord	Neutral Red				
O.V. 21	*M. smegmatis*	F	−	−	−	−	−	G
O.V. 87	*M. smegmatis*	F	−	−	−	−	−	FNF
O.V. 89	*M. phlei*	F	+	−	−	−	+	G
O.V. 119	*M.* spp.	F	−	−	−	−	−	FNF
O.V. 130	ATCC #4243	F	−	−	−	−	−	G
O.V. 628	"Radish" spp.	F	+	+	+	+	+	G
71	Skotochromogen spp.	W	−	−	−	−	−	FNF
98	Skotochromogen spp.	W	−	−	−	−	−	FNF
141	Skotochromogen spp.	W	−	−	−	−	−	FNF
201	*M. fortuitum*	W	−	−	−	−	−	G
208	*M. fortuitum*	W	−	−	−	−	−	G
209	*M. fortuitum*	W	−	−	−	−	−	G
287	*M. fortuitum*	W	−	−	−	−	−	G
9970	*N. asteroides*	ATCC	−	−	−	−	−	FNF

O.V.—Olive View accession numbers
G — greenish
FNF — Faint to no fluorescence
F — Froman; Olive View, California
W — Wayne, (Veterans Administration Hospital) California
ATCC (Washington)

TABLE 7

REACTIONS OF CLINICAL STRAINS OF M. TUBERCULOSIS* WITH AURAMINE METHODS

Organism	Total Strains	Auramine Disc +	Auramine Disc −	Auramine Flood +	Auramine Flood −	Cord +	Cord −	Auramine Fluorescence +	Auramine Fluorescence −
M. tuberculosis	43	43	0	43	0	43	0	Both G and YO types	0

* Arroyo Del Valle Sanatorium, Livermore, California
G—greenish
YO—yellow to orange

TABLE 8

AURAMINE FLOOD TEST VERSUS ROUTINE MICROSCOPIC EXAMINATION OF 100 CULTURE PLATES FOR M. TUBERCULOSIS

Reports on 100 Routine Cultures	Microscopic Colony Search Before Auramine Flood Test	Microscopic Colony Search After Auramine Flood Test
Positive for *M. tuberculosis*	43	46
Negative for *M. tuberculosis*	47	46
Contaminated cultures*	10	8
Total	100	100

* Contamination occupying approximately one-half or more area of the culture plate.

served in 10 strains and yellow to orange in 5 strains. Weak positive reactions with the neutral red test and the auramine disc test all exhibited greenish fluorescence.

V. Skotochromogen group: This group of 10 skotochromogens presented some difficulty with the auramine flood technique because of the deep orange color of the colonies. However, the test was modified as follows: A few drops of the reagent used in the auramine flood test were placed on the surface of the culture plate. The reaction was observed microscopically and the change in color, if any, was compared with neighboring colonies which served as controls. In the auramine disc test the color of the colonies did not appear to interfere, and the results paralleled the neutral red reaction in each case. Discrepancies were observed with strains Nos. 5, 15, and 190 by the auramine flood test. Cord formation was absent in this group of skotochromogens.

Fluorescence in this group fell into two categories: (*1*) greenish fluorescent strains; (*2*) faint to nonfluorescent strains. Yellow to orange fluorescence was not observed with this group of organisms.

VI. Miscellaneous acid-fast group: This group consisted of 14 various strains of mycobacteria, including one strain of *Nocardia asteroides*. There was again a definite correlation of results between the auramine (disc and flood tests) and the neutral red reaction. Organisms in this group were for the most part fast-growing saprophytes. The "radish" saprophyte was the only organism that was auramine positive and "corded." *M. phlei* was auramine negative and corded. "Cording" was absent in the remaining strains tested, including *Nocardia asteroides*.

As with the skotochromogen group, two categories of fluorescence were observed: (*1*) greenish fluorescent strains; (*2*) faint to nonfluorescent strains. The three skotochromogen saprophytes were faint to nonfluorescent types. *M. smegmatis* (O.V. 87), *M.* spp. (O.V. 119), and *Nocardia asteroides* exhibited similar behavior. The *M. fortuitum* strains uniformly exhibited a bright greenish fluorescence.

VII. M. tuberculosis group: Forty-three clinical strains of *M. tuberculosis* isolated from the sputum of 100 different patients were all positive by the auramine disc and flood tests. Debris or sediment carried over to these plates from the concentration procedure did not appear to interfere with the auramine flood-staining technique. Colonies stained uniformly yellow to bright yellow, leaving the surrounding debris and contamination unstained. "Cording" appeared to be present in all strains of this group and morphology was typical for *M. tuberculosis*.

Results with plates considered to be negative for tubercle bacilli: Because of the consistency with which the auramine flood test could disclose col-

onies of *M. tuberculosis* among the debris and contamination in routine laboratory cultures, it was decided to test the remaining negative and contaminated cultures by this method.

First, a routine microscopic search was made, which is the standard procedure used in this laboratory. The results of the examination were then reported as shown in table 8. Next, the plates were again examined microscopically after the addition of the auramine reagent.

The new method uncovered three additional auramine-positive cultures. Two were from cultures reported as contaminated and one from a culture reported as negative which harbored a small single colony among some debris which was not detected in the earlier microscopic examination.

Discussion

The auramine flood test is now routinely employed successfully in this laboratory on all cultures before discarding them as presumably negative. All contaminated plates are similarly tested. A positive auramine flood test would establish the presence of an auramine-positive strain of mycobacteria. A negative test would exclude the presence of *M. tuberculosis* colonies on the surface of the culture medium.

The auramine flood test has also been found very useful in drug-susceptibility testing. With primary isolation of *M. tuberculosis* from pathologic material on oleic-acid–albumin agar drug-containing media, contamination may obscure the reading on culture plates, thus making accurate drug-susceptibility determinations very difficult. The flood technique exposes the area of mycobacterial growth and permits a more accurate estimation of resistant colonies. Similarly, a debris-laden drug-susceptibility culture plate will expose hidden colonies by the flood technique and allow the technician to report susceptibility tests with a greater degree of accuracy. Doubtful susceptibility results will often require that that test be repeated, thereby resulting in valuable loss of time.

It is in these problems of contamination, false negative reports, and drug susceptibility determinations that the usefulness of the auramine flood test deserves emphasis.

It is believed that the auramine disc test will be most helpful as a rapid screening technique for routine cultures of mycobacteria and can certainly come within the scope of almost any clinical laboratory.

It was observed that the staining of mycobacterial organisms with auramine in the disc test and the subsequent application of an alkali probably measured the ability of the stained organisms to resist decolorization.

The nature of the reaction observed in the auramine flood test was not fully comprehended, and was investigated further.

It was noted that the test solutions could be added to the culture plates in any sequence without appreciably affecting the results. The sequences attempted in preliminary studies were as follows:

Alcoholic auramine flooding for one minute, followed by an alkali decolorization
Alkali flooding for one minute, followed by an alcoholic auramine solution
Combined alcoholic auramine and alkali solution (equal volumes) flooding for one minute

It was found that an auramine-positive colony could be stained equally well by any of the three methods described. From this observation it appeared probable that the alcoholic dye solution could penetrate the colonies more rapidly than the aqueous alkali solution under the same set of conditions. The cellular constituent of the colony involved in the reaction appeared to bind the dye, resulting in a brilliant yellow color extracted from a nearly colorless alkaline solution. Since auramine solutions were intensely yellow at neutral or acid pH, it was presumed that the reactive cellular constituent of the colony was acidic in nature. This supposition appeared to be substantiated by a quotation from the work by Richards (9):

> Of the many compounds isolated from tubercle bacilli by Professor Rudolph Anderson and his associates, only mycolic acid was found to be acid-fast. The auramine stained the mycolic acid intensely yellow at room temperature, and the combination resisted destaining in acid alcohol. On exposure to ultraviolet light an intense bright yellow fluorescence was observed.

It is believed that in the present preliminary series of auramine tests, mycolic acid appears to be the reacting cellular constituent in mycobacterial colonies. It was not established whether neutral red reacted with mycolic acid within the colony in the microcolonial neutral red reaction.

However, the parallel results obtained by two methods presented the possibility that an identical cytochemical reaction was taking place with both auramine and neutral red reagents.

In the auramine flood test it was observed that, after staining, colonies of *M. tuberculosis* were very difficult to decolorize by repeated alkali washes. However, preliminary experiments in this laboratory have indicated that eventually all types of mycobacteria would decolorize after sufficient exposure to alkali. It was also found that overexposure of colonies to alcohol in an alkaline solution could cause some types of auramine-positive mycobacteria to decolorize more rapidly. The age of the colonies was found to be another factor responsible for variable reactions. It was of interest to note that several refrigerated year-old cultures of *M. tuberculosis* were still able to produce a weak positive test by all three auramine methods.

In the auramine fluorescence test it was observed that miscellaneous acid-fast strains which were negative by the neutral red test and by the auramine disc and flood tests, were nevertheless positive by the fluorescence technique. This demonstrated that residual amounts of auramine dye (and possibly neutral red) remained bound in the colonies apparently undetected by the standard visual and microscopic techniques.

Several types of fluorescence were observed. The majority of mycobacterial strains fluoresced in the green spectrum, ranging from yellow green to blue green. It was not always possible to visually distinguish between these various types. Three categories of fluorescence could be sufficiently well segregated: greenish, yellow to orange, and faint to nonfluorescent. The third category was interesting from the standpoint that four skotochromogens exhibited this phenomenon. These were positive by both the neutral red and the auramine disc tests. Similarly, a group of three skotochromogen saprophytes which were negative by both the neutral red and the auramine disc and flood tests showed faint to nonfluorescent reactions.

In order to exclude the faint to nonfluorescent phenomenon as perhaps due to the amount of test material present on the disc, duplicate discs were tested with light and heavy impregnations of the same organism. It was found that heavily impregnated discs did not significantly reveal an increase in fluorescence.

The significance of the three categories of fluorescence was not investigated further, but it was believed that this phenomenon was apparently dependent on the quantity of a reactive constituent present in the body of the mycobacteria which was postulated to be mycolic acid.

In conclusion, the auramine disc test eliminated the necessity of making routine smears from suspected colonies, since only the auramine-positive strains of acid-fast mycobacteria were visualized in the test. The procedure was relatively simple, rapid, and accurate, with results essentially comparable with the neutral red reaction by the microcolonial technique.

Summary

A new reagent has been found (alkaline auramine) which differentially stains mycobacterial colonies brilliant yellow *in situ*.

In preliminary studies, 59 strains of atypical mycobacteria and 43 clinical strains of *M. tuberculosis* were tested by three auramine techniques: the auramine disc test, the auramine flood test, and the auramine fluorescence test.

Auramine is a fluorescent dye, and experiments appeared to indicate that, when masses of mycobacteria were treated with this dye, they fluoresced under an ultraviolet blacklight lamp, but with a color spectrum depending apparently on the quantity of a reactive constituent present in the body of the mycobacteria which was postulated to be mycolic acid.

The results with the auramine disc and flood tests appeared to parallel the neutral red reaction by the microcolonial method.

Sumario

Rápida Identificación de las Colonias de Micobacterias. I. La Prueba de la Auramina

Se ha descubierto un nuevo reactivo (la auramina alcalina) que tiñe diferencialmente de color amarillo brillante las colonias de micobacterias *in situ*. En estudios preliminares, se comprobaron 59 cepas de micobacterias atípicas y 43 cepas clínicas del *M. tuberculosis* con tres técnicas de la auramina: prueba del disco de auramina, prueba del baño de auramina y prueba de la fluorescencia de auramina.

La auramina es un colorante fluorescente y los experimentos parecían indicar que, cuando se tratan con ella masas de micobacterias, éstas muestran fluorescencia bajo la luz negra de una lámpara de rayos ultraviolados, pero con un espectro de colores que dependen al parecer de

la cantidad de un componente reactivo que haya en el cuerpo de las micobacterias, que se presupone es ácido micólico.

Los resultados obtenidos con las pruebas del disco y del baño de auramina parecían ser paralelos a los de la reacción del rojo neutro con el método microcolonial.

Resume

Identification rapide de colonies de mycobacteries. Par le test à l'auramine

Un nouveau réactif a été decouvert (auramine alcaline) qui différencie les colonies de mycobactéries par leur couleur: un jaune brillant et ceci *in situ*.

Des études anterieures, 59 souches de Mycobactéries atypiques et 43 souches de *M. tuberculosis* furent etudiées avec trois techniques utilisant l'auramine : la méthode du disque, la méthode d'immersion et la méthode de fluorescence.

L'auramine est une matière colorante et les etudes experimentales montrent que, lorsqu' une quantité massive de Mycobactéries est traitée avec cette matière, elle devient fluorescente en lumiere noire, mais avec un spectre coloré dependant, semble-t-il, de la quantité d'une substance qui réagit et qui est présente dans les corps des bactéries. Cette substance est, pense-t-on, de l'acide mycolique.

Les résultats avec les techniques du disque et d'immersion semblent parallèles à ceux obtenus par la réaction au rouge neutre, appliquée a des microcolonies.

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MODIFICATION OF EXPERIMENTAL MURINE TUBERCULOSIS BY AN ANTIMICROBIAL AGENT, STREPTOVARICIN[1]

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(Received for publication July 7, 1959)

INTRODUCTION

It is generally recognized that the white mouse possesses greater resistance than most mammalian species to tuberculous infection and, furthermore, that the histopathologic response of this species is characterized by distinctive dissimilarities. According to Pagel (1), "the characteristic feature in mice is the absence of caseation, giant cells and tubercles. In their stead, however, is the formation of foam cells and within them the acid-fast bacilli persist for a long time." In addition to an extensive review of the literature, Raleigh and Youmans (2, 3) made a comprehensive study of the pathology and pathogenesis of the experimental infection in white mice. The latter report has given adequate basis for the pathologic interpretation of the efficacy of antimicrobial agents in the suppression of tuberculous lesions in mice. These writers recognized that the preeminent pulmonary changes of this infection were of two general types, "proliferative" and "necrotic-exudative"; these types they regarded as associated with low and high velocity evolution, respectively, of tuberculous involvement.

The antituberculous effectiveness of streptovaricin[2] (4) in fulminant murine infection has been given in earlier reports (5, 6). These *in vivo* studies have shown that treatment for 10 days with a drug concentration of streptovaricin which afforded 100 per cent survivors at the end of a 28-day postinfective period resulted in greater survival time during a total period of 84 days than in similarly infected groups of mice treated with comparable dosages of isoniazid and streptomycin. From data obtained by the microbial enumeration technique, McCune and associates (7) regarded streptovaricin as having only a low degree of effectiveness in chronic tuberculous infection in the mouse during the first sixty days of therapy, but the drug had more effectiveness during the third month of its administration. The following study was undertaken, therefore, to determine the nature of pathologic and bacteriologic changes in experimental murine tuberculosis subjected to streptovaricin therapy of various durations and to elucidate the nature of the prolonged survival effect in the mouse associated with the administration of this antimicrobial agent.

METHODS AND MATERIALS

CF_1 male mice weighing 16 to 18 gm. were used. *Mycobacterium tuberculosis* var. *hominis* (H37Rv) was maintained on Löwenstein agar slants in the manner previously described (6). The infection was established by injection of 0.5 ml. of culture (approximately 6.8×10^6 viable units of tubercle bacilli) in the tail vein; treatment when indicated was initiated within three hours. Streptovaricin was administered by oral intubation using 0.5 ml. of the drug suspended in 2.5 per cent gelatin in physiologic saline.

Several hundred mice were infected as described above and were divided into groups to receive the following treatment: *Group A* was treated daily with 400 mg. of streptovaricin for 152 days, whereas *Groups B* and *D* received the same daily dosage for ten and seventy days post infection, respectively. *Group C* mice were infected and untreated. Mice were killed for preparation of lung and spleen tissue homogenates (3 mice) and for pathologic examinations (3 mice) at twenty intervals during the course of the study as follows: 1, 2, 5, 7, 9, 12, 14, 16, 21, 27, 35, 40, 47, 77, 87, 100, 113, 128, 142, and 152 days post infection. The untreated group was not examined after the sixteenth day post infection.

The preparation of tissue homogenates and viable unit determinations were essentially as described by McCune and Tompsett (8). The method for the determination of the end points of the *in vitro* susceptibility data has also been previously described (4).

The brain and thoracic, and abdominal and pelvic, viscera were fixed in Bouins fluid. These tissues were embedded in paraffin, sectioned at 8 μ and stained by the Harris' hematoxylin-eosin method and by Kinyoun's method for the demonstration of acid-fast bacilli.

[1] From the Research Laboratories, The Upjohn Company, Kalamazoo, Michigan.

[2] The trade name of The Upjohn Company for streptovaricin is Dalacin.

Results

Bacteriologic Findings

Bacteriologic examination of the lungs (figure 1) and spleen (figure 2) revealed that the viable cell population rose rapidly to more than 10^8 cells (*Group C*). In contrast, the animals treated with streptovaricin showed a marked reduction in the microbial population of both organs. Continuous use of streptovaricin throughout

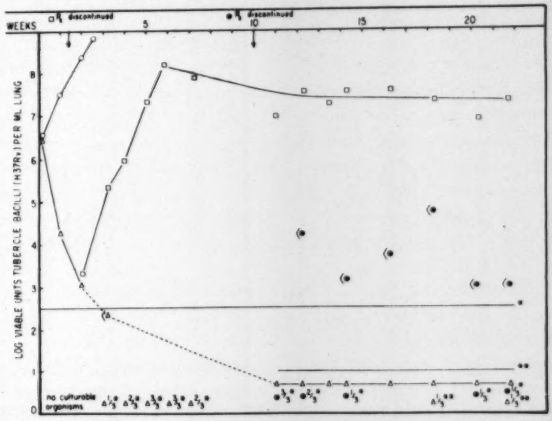

FIG. 1. Effect of different durations of streptovaricin therapy (400 mg. per kg. per day orally) on populations of tubercle bacilli (H37Rv) in mouse lungs.

○, Control; △, 152 days; □, 10 days; ●, 70 days.

* The technique which was used permitted detection of 70 to 90 culturable units of tubercle bacilli per lung.

** The technique which was used in *Group A* (△), from the seventy-seventh day on permitted detection of one to three culturable units of tubercle bacilli per lung.

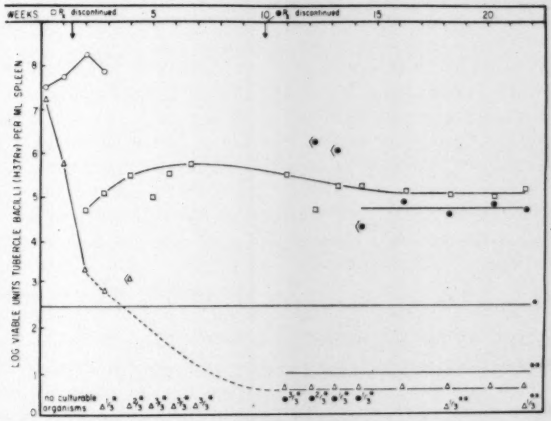

FIG. 2. Effect of different durations of streptovaricin therapy (400 mg. per kg. per day orally) on populations of tubercle bacilli (H37Rv) in mouse spleens.

○, Control; △, 152 days; □, 10 days; ●, 70 days.

* The technique which was used permitted detection of 70 to 90 culturable units of tubercle bacilli per spleen.

** The technique which was used in *Group A* (△), from the seventy-seventh day on, permitted detection of one to three culturable units of tubercle bacilli per spleen.

the experiment (*Group A*) resulted in effective control of tubercle bacilli, as evidenced by the extremely low mycobacterial population in these organs. Interruption of therapy at ten days post infection (*Group B*) was followed by a rapid rise in the population of acid-fast bacilli in both lungs and spleen, indicating that streptovaricin failed to sterilize these tissues. The microbial flora in these organs was comparable with that observed in the infected untreated mice but, unlike the results obtained with the infected untreated controls, none of the animals dosed with streptovaricin died. It may also be seen that 70 consecutive days of treatment with streptovaricin (*Group D*) did not sterilize the lungs and spleen although the termination of therapy at this point did not result in the rapid increase in organ population observed in the period following ten days of streptovaricin treatment and subsequent interruption of therapy.

Tubercle bacilli isolated from *Group A* mice after 93 days of antimicrobial therapy showed *in vitro* susceptibility to streptovaricin; this culture also was found to be virulent for normal CF_1 mice. However, organisms which were recovered from the one hundred and twenty-eighth day on showed *in vitro* resistance to more than 10γ of streptovaricin per ml.

Gross Pathologic Findings

Untreated Group C: Slight hypertrophy of the spleen was first observed on the fifth day post infection; by the ninth day splenomegaly was well developed. Focal subpleural lesions (1 mm. or less in diameter) in the lungs were first detected on the twelfth day of infection; a few subepicardial lesions were also seen. Until observations were terminated at 16 days, the lung foci became more numerous and showed coalescence. Occasionally small subpleural hemorrhages were seen.

Treated Group A (152 days): Mice of this group also showed slight splenic hypertrophy on the fifth day post infection; however, maximal enlargement, which appeared to occur later than in untreated mice, was seen at 12 days. After 21 days post infection and during the remainder of the study, enlargement of the spleen became less obvious except for a brief period 113 to 128 days post infection when hypertrophy again was noticed in some mice.

At 21 days, random whitish foci, 1 to 2 mm. diameter, were seen in the liver. Noticeable body weight gains were observed in this group of mice at this time. A few focal lung lesions were seen in one mouse 142 days post infection.

Treated Group B (10 days): Splenic hypertrophy became noticeably greater than in *Group A* mice 21 days post infection; this finding was constant thereafter.

From the twenty-seventh day, focal lung lesions were seen with increasing frequency. Confluent lesions became prominent 47 days post infection, and at subsequent observations the lungs of these mice usually revealed relatively large areas of consolidation. Mice of this group were appreciably smaller than *Group A* mice after the twenty-seventh day of infection. At 142 days, the kidneys showed grayish subcapsular discoloration.

Treated Group D (70 days): Splenomegaly became apparent on the 128th day, about two months after treatment was discontinued.

A few subpleural foci were seen in the lungs of a single mouse 142 and 152 days post infection.

Histopathologic Findings

Untreated Group C: One day post infection, numerous randomly distributed inflammatory foci, 100 to 300 μ in diameter, were present in the liver and lungs; most of the acid-fast bacilli appeared to be contained by polymorphonuclear phagocytes.

Conspicuous hyperplasia of reticuloendothelial cells was shown in the spleen on the fifth day. Seven days post infection, the primary reactive foci in the liver were enlarged, 300 to 500 μ in diameter, and numerous secondary foci were present. The cellular increment of these foci consisted chiefly of lymphocytes and mononuclear phagocytes. Widespread necrosis of hepatic cells was also evident.

Progressive inflammatory involvement was shown in several organs by the ninth day of the infection. The impression was obtained that the liver, spleen, kidneys, heart, and probably gastric mucosa contained numbers of acid-fast bacilli comparable with those in the lungs. The definite increase of megakaryocytes in splenic sections was regarded as a reflection of hematopoietic response during this stage of the infection.

Observations on the twelfth day revealed a noticeable increase in organisms and inflam-

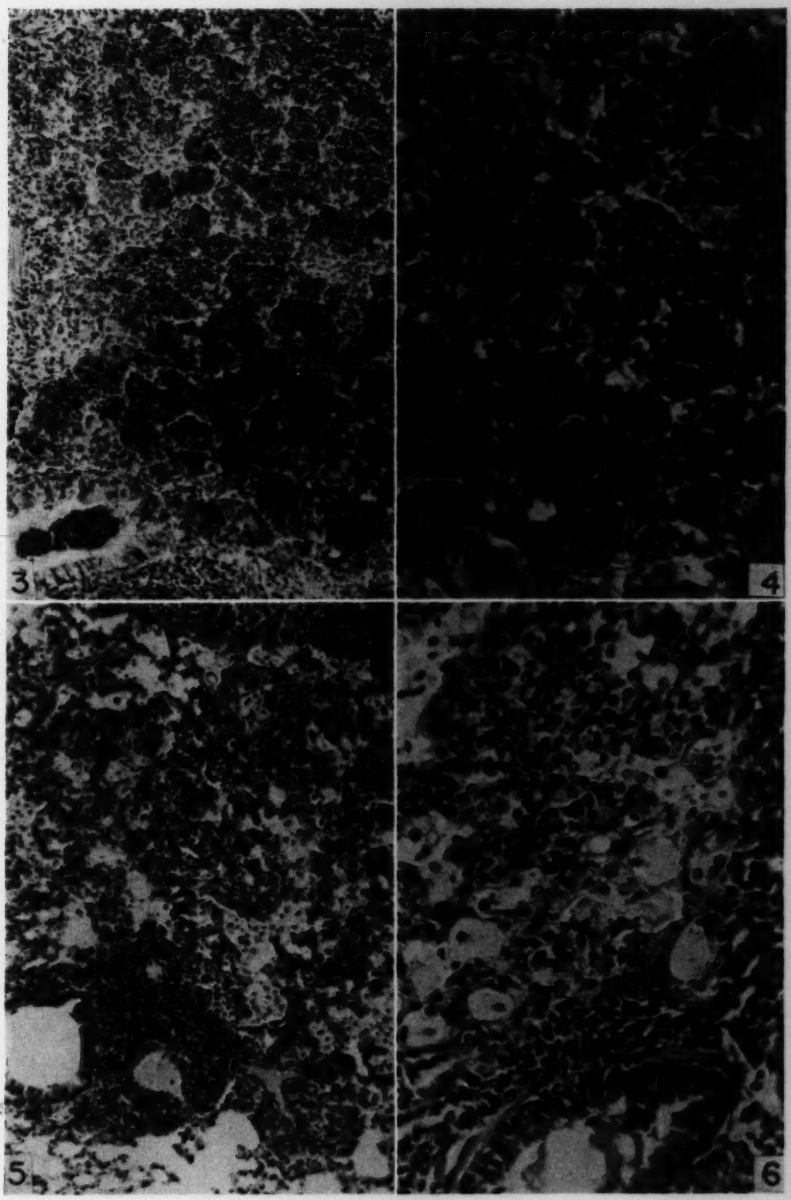

FIG. 3. Fulminant murine tuberculosis in the lung 16 days post infection (*Group C*). (× 120)

FIG. 4. Enlargement of figure 3, showing masses of degenerating cellular exudate, chiefly polymorphonuclear phagocytes, in the distended alveolar spaces. (× 290)

FIG. 5. Inflammatory response 113 days post infection in the lung of a mouse which received streptovaricin therapy for ten days (*Group B*). (× 120)

FIG. 6. Enlargement of figure 5. Foam-cell phagocytes in the alveolar spaces and interstitial infiltration of lymphocytes are shown. The presence of these cells appeared to be associated with host resistance to tuberculous infection. (× 290)

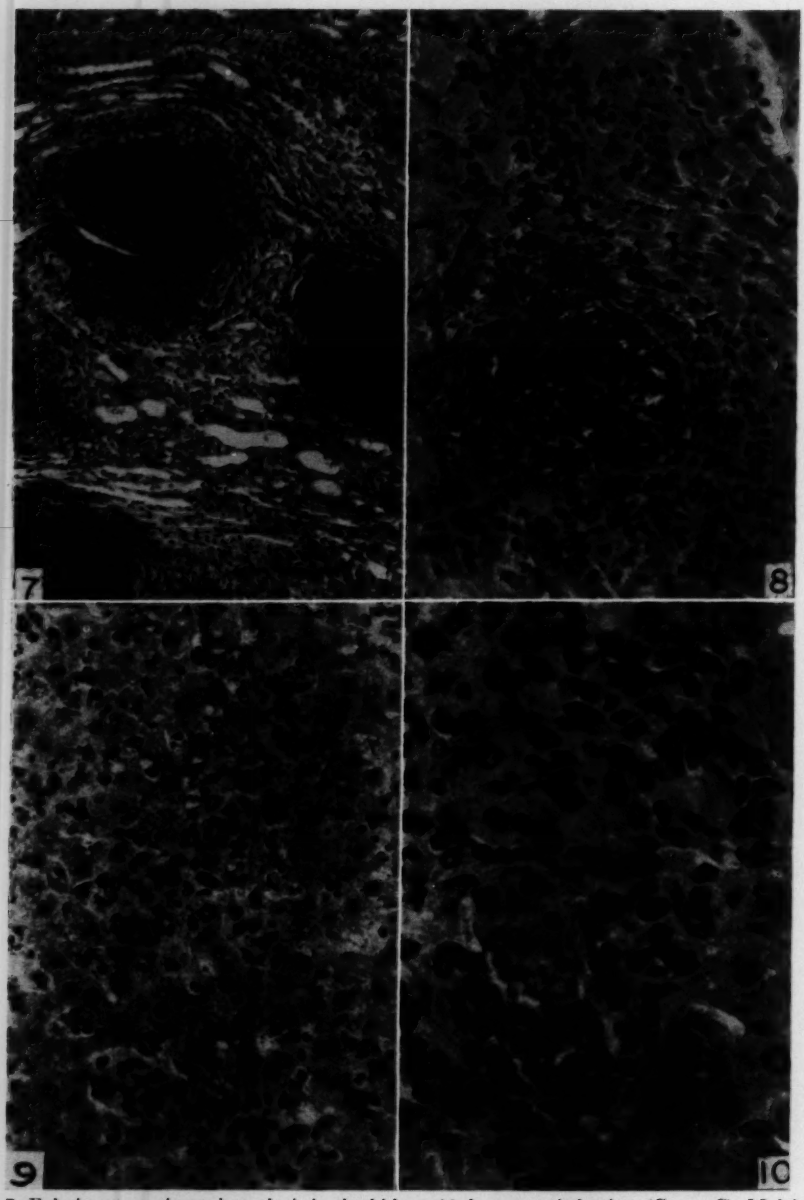

FIG. 7. Fulminant murine tuberculosis in the kidney 16 days post infection (*Group C*). Multiple abscesses are shown in the renal medulla. (× 100)

FIG. 8. Photomicrograph of mouse heart 12 days post infection showing necrotic tuberculous focus and mixed inflammatory response in the myocardium (*Group C*). (× 290)

FIG. 9. Diffuse and nodular inflammatory response of mononuclear phagocytes and lymphocytes shown in a liver section 16 days post infection (*Group C*). (× 290)

FIG. 10. Perivascular nodule of inflammatory cells, in which epithelioid cells are prominent, in the liver of a *Group B* mouse 40 days post infection. (× 570)

matory cells in the lungs, predominantly mononuclear phagocytes. Areas of pulmonary consolidation, approximately 0.5 mm. in diameter, resulted from the enlargement of earlier septal inflammatory foci together with the collapse of adjacent alveoli. An accelerated extension of lesions also occurred in the kidneys (figure 7) and heart (figure 8).

The pulmonary foci showed a remarkable transition about 14 days post infection from a predominantly interstitial reaction of mononuclear phagocytes to alveolar plugging with a preponderance of polymorphonuclear phagocytes. A marked increase in acid-fast bacilli was found in the degenerating alveolar exudate.

Terminal observations on the lung 16 days post infection showed more severe manifestations of the fourteenth-day findings as a result of extension and coalescence of the "necro-exudative" foci (figures 3 and 4). The centers of these foci showed coagulation necrosis and some loss of pulmonary framework. The liver was diffusely involved by a further increase of small collections of lymphocytes and mononuclear phagocytes; some of the latter were epithelioid in type (figure 9).

Treated Group A (152 days): The distribution of inflammatory foci and acid-fast bacilli in several organs was similar to that in the untreated group one day post infection.

After one week of treatment, considerable deceleration in the development of lesions and proliferation of acid-fast bacilli was observed. The spleen showed well-developed hyperplasia of reticuloendothelial cells. The liver revealed an inflammatory transition from the earlier random distribution of paravascular foci to chronic perivascular collections of lymphocytes and mononuclear phagocytes.

Chronic tuberculous foci were found in several organs from the sixteenth to the twenty-seventh days post infection. The liver showed the most extensive inflammatory reaction and contained most of the relatively few acid-fast bacilli which were seen. Small glial foci and perivascular infiltration of mixed inflammatory cells were seen in the brain stem and cerebrum.

The most remarkable finding from 35 days post infection to the termination of the study was the presence of epithelioid cells in the centers of the small nonprogressive tuberculous foci, particularly in the liver; acid-fast bacilli were present in these foci.

Treated Group B (10 days): From 35 to 87 days post infection, there was a noticeable reactivation of the infection in the lungs and liver and, to a lesser degree, in the kidneys, heart, pancreas, and brain. A significant increase of acid-fast bacilli was seen in tuberculous lesions and, rather uniformly, epithelioid cells appeared in these foci 35 to 40 days post infection (figure 10). In the lungs, particularly, tuberculous foci in the alveolar walls were enlarged and more numerous. In the more rapidly developed foci, collapse of the alveolar walls and an influx of large foam cells into the alveolar spaces were seen; some sections showed more than 50 per cent involvement 87 days post infection. The lymph nodes, especially the tracheobronchial nodes, showed marked proliferation of reticuloendothelial cells and a few foci of epithelioid cells; acid-fast bacilli were fairly common in these nodes. Similarly, accelerated reticuloendothelial hyperplasia was shown in the spleen.

After 100 days post infection, a continual influx of lymphocytes contributed to the diffuse tuberculous involvement in the lungs (figures 5 and 6). The alveolar exudate of foam cells became increasingly abundant. Examination of acid-fast stained preparations indicated an appreciable reduction of organisms during this period of increased inflammatory response.

Treated Group D (70 days): Thirty days after treatment was discontinued the mild involvement of tissues from this group of mice still resembled the residual tuberculous foci in *Group A* mice. The major tissue alteration was observed toward the termination of the study; about 142 days post infection focal lymphoid hyperplasia appeared in the lungs and kidneys similar to that observed earlier at about 100 days in *Group B* mice. Acid-fast bacilli were detected in some of these foci.

Discussion

The remarkable effectiveness of streptovaricin in the suppression of fulminant murine tuberculosis, which had been demonstrated in earlier studies (5, 6) by the prolonged survival time of treated mice, has been more fully substantiated by the results of microbial enumeration data of the lung and spleen and by the histopathologic findings. For the most part, valid correlations could be made between the extent of the pathologic processes and the number of culturable

organisms in the lung and spleen. Similar to the experience of McCune and Tompsett (9), the degree of correlation became somewhat lessened during the recrudescent stage of the infection in *Group B* mice, during which the tubercle bacilli census was stabilized. It is of further interest, since these observations were made by different investigators in the present study, that the relative abundance of acid-fast bacilli observed microscopically in the tissue sections compared favorably with the viable cell enumeration data. A notable exception occurred in which visual reduction of acid-fast bacilli in the lungs was not particularly apparent from the viable cell enumeration data obtained after 100 days post infection in *Group B* mice.

It has been well recognized (10) by both pathologic and bacteriologic criteria that the predominant changes in murine tuberculosis occur in the lung. From the comparison afforded by the data of the various groups of the present study, it was evident that the lung was subjected to a greater range in number of organisms and in extent of the inflammatory response than the spleen. In the untreated mice of *Group C*, progressive tuberculous lesions were observed in several organs, including the lung, until about the twelfth day post infection, when morphologic evidence of breakdown of host resistance appeared in the lung, and the terminal necrotic-exudative changes set in (figures 3 and 4). In the liver and spleen the inflammatory response remained progressive and largely focal until observations were discontinued; there were no survivors 19 days post infection (figure 9). The terminal increase in acid-fast bacilli and reactive foci of inflammatory cells in the liver may have been sequential to the shedding of organisms into the circulation as a result of the breakdown of pulmonary resistance to tuberculous infection. The mortality which ensued presumably could be associated in large measure with the widespread involvement in the lung after the twelfth day of the fulminant infection. In addition, the scattered areas of necrosis and abscess formation in the heart (figure 8) and kidneys (figure 7) could not be disregarded as contributing factors in this respect.

The results in the *Group A* mice which received an equivalent of 400 mg. of streptovaricin per kg. daily during the study were characterized by rapid decrease of tubercle bacilli to below 70 to 90 culturable units per lung or spleen during the first two or three weeks of treatment. This decrease in microbial population was more rapid than that experienced by McCune and associates (7) and is undoubtedly explained by differences in drug administration. It should be recalled that streptovaricin was administered in the present experiments by oral intubation, whereas in the studies of McCune and associates the drug was incorporated in the diet. It is pertinent that the daily regimen of streptovaricin did not eliminate all tubercle bacilli from the lung and spleen. One to three culturable units were detected in some mice of this group from the seventy-seventh day until the study was terminated. *In vitro* resistance to streptovaricin of tubercle bacilli recovered from this group was not demonstrated, however, prior to the one hundred and twenty-eighth day of treatment. The host response of *Group A* mice from 35 to 152 days post infection was characterized by the presence of small epithelioid nodules, particularly in the liver (figure 10). The distribution of these nodules was closely associated with the presence of persistent tubercle bacilli which could be seen in the tissues. It must be concluded, then, that dormant infection remained in the continuously treated *Group A* mice for the entire period of the study.

Following the discontinuation of streptovaricin therapy in *Group B* mice after the tenth day post infection, rapid multiplication of the remaining tubercle bacilli occurred. In the lung, the log of viable units rose from approximately 10^3 to 10^8 at a rate comparable with that of the untreated *Group C* mice. The multiplication in the spleen after therapy was appreciably less, by comparison, and the census of tubercle bacilli in this organ became stabilized between 10^5 and 10^6 for the remainder of the study. The development of the host inflammatory response to the reactivated infection appeared to lag but became synchronously noticeable between the fifth and sixth weeks of the study with the abrupt leveling of the tubercle bacilli census in the lung (figure 1). A plateau in the enumeration data was shown for the duration of the study, although at different levels of growth population, in the lungs and spleens of this group of mice. Histologically, during this period, the pulmonary alveoli became progressively involved with inflammatory exudate predominantly composed of phagocytic foam cells in which most of the tubercle bacilli appeared to be

contained. As suggested by Pagel (1), it likewise seems reasonable from these findings that the appearance of foam cells was closely associated with the specific host resistance of the mouse. This tissue reaction, as described in the pathologic results, corresponded to the "proliferative" type of reaction of Raleigh and Youmans (3) and was distinct from their "necrotic-exudative" type which was seen in the fulminant tuberculous infections of untreated *Group C* mice.

About 100 days post infection the inflammatory response in the lungs of *Group B* mice was further expanded by an influx of lymphocytes, particularly in the perivascular and peribronchial areas. The probable immunologic function of this nonphagocytic cell has long been considered. Dubos (10) has emphasized that tuberculous immunity can develop only when an adequate amount of antigen is present in the tissue. In *Group B* mice this was undoubtedly provided by the extensive multiplication of tubercle bacilli after discontinuation of streptovaricin therapy on the tenth day. Significantly, however, the same extensive multiplication of cells after termination of isoniazid or streptomycin therapy (6) did not result in a prolonged survival rate comparable with that of streptovaricin-treated mice. Thus, a morphologic basis for understanding the prolonged survival time of mice of this group may reside in the phagocytic response of the foam cell and in the probable antibody-forming property of the lymphocyte (figures 5 and 6). No explanation for the delayed appearance of the lymphocytic infiltration in the lung, more than two months after the population of tubercle bacilli became stabilized, was obtained. It is noteworthy, however, that a similar response in *Group D* mice, which were treated for 70 days, did not appear until the observations were made at 142 days or after a comparable period of more than two months. Although any significant degree of mortality in the treated groups was lacking, it should not be overlooked that the extensive pulmonary involvement of *Group B* mice at the terminal observations strongly suggested that the survival time of these mice beyond 152 days was limited.

Summary

The effect of the antimicrobial agent streptovaricin on fulminant murine tuberculosis was evaluated by parallel studies of microbial enumeration in the lung and spleen and histopathologic interpretation.

In one group of mice, daily oral intubation of an equivalent of 400 mg. of streptovaricin per kg. effectively suppressed the infection. Complete eradication of the infecting organism was not obtained from the seventy-seventh day to the termination of the study; one to three culturable units of tubercle bacilli were detected in some mice of this group.

The infection was rapidly reactivated in a second group when a comparable dosage of streptovaricin was limited to ten days post infection. In contrast to the results expected from the rapid rise in viable cells, the mortality did not differ from that of the group receiving treatment over a period of 152 days. The development of host resistance was regarded as responsible for the prolonged survival time of mice of this group for more than 152 days. Morphologically, the host response was associated with the presence of lymphocytes and phagocytic foam cells in the lung.

A slight reactivation of the tuberculous infection was seen terminally in a third group of mice which were dosed at 400 mg. per kg. for 70 days.

Sumario

Modificación de la Tuberculosis Murina Experimental por un Antimicrobiano, la Estreptovaricina

Por medio de estudios paralelos de la numeración microbiana en el pulmón y el bazo y de la interpretación histológica, se justipreció el efecto del agente antimicrobiano, la estreptovaricina, sobre la tuberculosis murina fulminante.

En un grupo de ratones, la intubación oral diaria de un equivalente de 400 mgm. de estreptovaricina por kg. de peso suprimió eficazmente la infección. No se obtuvo la erradicación total del microbio infectante desde el septuagésimo-séptimo día hasta la terminación del estudio, descubriéndose de una a tres unidades culturables de bacilos tuberculosos en algunos ratones de este grupo.

La infección se reactivó rápidamente en un segundo grupo al limitarse una dosis comparable de estreptovaricina a los diez días consecutivos a la infección. En contraposición a los resultados esperados a la luz del rápido aumento de las células viables, la mortalidad no discrepó de la del grupo que recibió tratamiento durante un período de 152 días. La formación de resistencia en el huésped se consideró como causante del prolongado tiempo de sobrevivencia en los ratones

de este grupo a más de 152 días. Morfológicamente, la respuesta del huésped se asoció con la presencia de linfocitos y de glóbulos xantomatosos fagocíticos en el pulmón.

En un tercer grupo de ratones que recibieron 400 mgm. por kg. durante 70 días, se observó al terminar el estudio leve reactivación de infección tuberculosa.

Resume

Modification de la tuberculose expérimentale de la souris par un agent antimicrobien: La streptovaricine

L'effet anti-microbien de la streptovaricine sur la forme foudroyante de tuberculose de la souris a été jugée par des études portant parallèlement sur la numération des germes dans les poumons et la rate, et par l'interprétation histopathologique.

Chez un groupe de souris, il a été donné per os l'équivalent de 400 mg de streptovaricine par kilogramme, ce qui a supprimé l'infection. Une complète destruction du germe infectant n'a pas été obtenue de 77ème jour jusqu'à la fin de l'expérience. Du un à trois specimens de bacilles tuberculeux ayant pu être cultivés, furent découverts chez des souris de ce groupe.

L'infection fut rapidement réactivée dans un second groupe quand la même dose de streptovaricine fut limitée a 10 jours après l'infection. Contrairement au resultat que l'on pouvait attendre de l'augmentation rapide des germes vivants, la mortalité n'a pas été différente de celle du groupe ayant été traité pendant une periode de 152 jours. Le développement d'une résistance de la part de l'hôte est considerée comme responsable de la plus longue durée (plus de 152 jours) de vie des souris de ce groupe. Morphologiquement, la résistance de l'hôte est associée à la présence de lymphocytes et de cellules phagocytes dans le poumon.

Une légère réactivation de l'infection a été notée chez un troisième groupe de souris qui reçurent 400 mg par kilogramme pendant une période de 70 jours.

Acknowledgment

The writers wish to acknowledge the technical assistance of H. Kurtz, J. Kay, and D. Johnson, and the help of the photographic section in the preparation of illustrations.

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RESULTS OF THERAPY WITH THIOCARBANIDIN USED WITH ISONIAZID OR STREPTOMYCIN[1, 2, 3]

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(Received for publication June 10, 1959)

Introduction

Thiocarbanidin, a thiocarbanilide derivative, was found to have antituberculous activity *in vivo* and *in vitro* by Youmans and co-workers (1). *In vitro*, the activity in modified Proskauer and Beck medium was of approximately the same degree as streptomycin, but the antibacterial activity was appreciably reduced following the incorporation of serum into the medium. In a very low dosage, thiocarbanidin produced a significant prolongation of the survival of tuberculous mice. Youmans and associates stated that the chemotherapeutic effect of thiocarbanidin did not increase markedly with large increases in the drug intake and, at low dosage, the median survival time of mice was increased to the same extent as observed with the largest doses of PAS. Their results suggested that thiocarbanidin might be used in combination with streptomycin or isoniazid in place of PAS for the treatment of tuberculous patients. The suggestion was made from the experimental data that less than 1 gm. of thiocarbanidin might be substituted for 12 gm. of PAS.

Steenken and associates (2) showed that 0.5γ per ml. of thiocarbanidin produced inhibition *in vitro* of growth of tubercle bacilli resistant to isoniazid and to PAS in four freshly isolated strains of tubercle bacilli, but that tubercle bacilli resistant to amithiozone were resistant to 50γ of thiocarbanidin per ml. Thiocarbanidin-resistant strains of bacilli could be produced by serial transfers in medium containing increased concentrations of the drug. When thiocarbanidin was used to treat tuberculous guinea pigs, it was found to be most effective in combination with streptomycin or isoniazid. Isoniazid alone, however, was more effective than streptomycin and thiocarbanidin but not quite so effective as isoniazid and thiocarbanidin. Thiocarbanidin was found to be nontoxic for rabbits and guinea pigs.

Dill and Glazko (3) determined that the plasma concentrations of thiocarbanidin administered orally were quite low in humans and animals, ranging from a few tenths of a microgram to several micrograms per milliliter and that the urinary excretion accounted for only a few tenths of one per cent of the dose. The fecal excretion of the drug given orally was high, indicating poor intestinal absorption.

Phillips (4) treated twelve tuberculous patients with thiocarbanidin and isoniazid or streptomycin, and four patients with thiocarbanidin, streptomycin, and isoniazid. He confirmed the results of low absorption, slow excretion, and high fecal concentrations of thiocarbanidin, in addition to confirming the lack of toxicity to a dosage of 1 gm. of thiocarbanidin daily. Too few patients were present in the study for evaluation of the therapeutic efficacy of thiocarbanidin.

Method of Study

A study involving patients with active pulmonary tuberculosis was set up in certain Veterans Administration hospitals to test the therapeutic efficacy of thiocarbanidin in combination with streptomycin or isoniazid, while therapy with PAS and streptomycin or isoniazid was used for comparison. The study also sought to determine the efficacy of thiocarbanidin in delaying the emergence of bacterial resistance to the companion drug. All patients had sputum containing tubercle bacilli at the time of the study or had had similarly infectious sputum reported shortly before admission to the study. The study began in most hospitals in July, 1958, in previously untreated and re-treatment cases, and the analysis of final results extended to April 1, 1959, for those

[1] From the Pulmonary Disease Service, Veterans Administration Medical Teaching Group Hospital (Kennedy), Memphis, Tennessee.

[2] Presented, in part, at the Eighteenth Conference on the Chemotherapy of Tuberculosis, Veterans Administration–Armed Forces, St. Louis, Missouri, February, 1959.

[3] The study on thiocarbanidin was a protocol study for the chemotherapy of tuberculosis in the following Veterans Administration hospitals: Brecksville, Ohio; Buffalo, New York; Livermore, California; Memphis, Tennessee; Oteen, North Carolina; Sunmount, New York; and Whipple, Arizona.

patients still in the study. The particular drug regimen which the untreated and re-treatment patients received was determined by random allocation using a sealed envelope system. In one hospital, in the previously untreated group only, streptomycin was used with either thiocarbanidin or PAS, while in all of the other hospitals, isoniazid was used with thiocarbanidin or PAS. Among the re-treatment patients, thiocarbanidin or PAS was used with either streptomycin or isoniazid in accordance with recent drug-susceptibility studies for streptomycin or isoniazid. The previously untreated patients received either 1 gm. or 0.5 gm. of streptomycin daily, or 300 mg. of isoniazid daily, and 1 gm. of thiocarbanidin daily in a single dose, or 12 gm. of PAS daily. The re-treatment patients received 1 gm. of thiocarbanidin or 12 gm. of PAS daily with either 300 mg. of isoniazid or streptomycin daily. The re-treatment patients were given streptomycin daily in a dose of 1 gm. or 0.5 gm., or 1 gm. twice weekly.

In the previously untreated group, 23 patients received isoniazid and thiocarbanidin, 24 patients received isoniazid and PAS, 7 patients received streptomycin and thiocarbanidin, and 7 patients received streptomycin and PAS (table 1). More than half of the patients who received isoniazid with PAS or thiocarbanidin were treated for longer than four months, and approximately half of the group which received streptomycin received at least four months of therapy. In the re-treatment group, 18 patients received isoniazid and thiocarbanidin, 7 received isoniazid and PAS, 15 had streptomycin and thiocarbanidin, and 6 had streptomycin and PAS (table 2). Among the re-treatment patients, less than 50 per cent of the patients who received each drug combination had more than four months' therapy except the group of patients who received streptomycin and PAS, among whom 50 per cent were treated for more than four months.

TABLE 1

NUMBER OF PREVIOUSLY UNTREATED PATIENTS STUDIED

Regimen	Duration of Therapy				
	Less than 2 Months	2-4 Months	4-8 Months	More than 8 Months	Total
Isoniazid-thiocarbanidin	1	5	17	0	23
Isoniazid-PAS	4	6	11	3	24
Streptomycin-thiocarbanidin	0	4	2	1	7
Streptomycin-PAS	1	2	2	2	7

TABLE 2

NUMBER OF RE-TREATMENT PATIENTS STUDIED

Regimen	Duration of Therapy				
	Less than 2 Months	2-4 Months	4-8 Months	More than 8 Months	Total
Isoniazid-thiocarbanidin	3	7	5	3	18
Isoniazid-PAS	1	3	3	0	7
Streptomycin-thiocarbanidin	2	7	3	3	15
Streptomycin-PAS	3	0	3	0	6

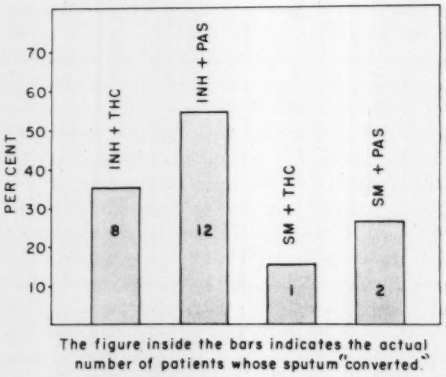

FIG. 1. The number and percentage of previously untreated patients who had reversal of infectiousness are demonstrated according to the therapy given.

RESULTS

Reversal of infectiousness: The disappearance of tubercle bacilli from the respiratory secretions, as determined by microscopy or culture for at least two months, occurred to the greatest extent in the previously untreated groups and among those patients who received isoniazid. The percentage with reversal of infectiousness is shown in figure 1. Those patients whose sputum was infectious only prior to hospitalization were eliminated from consideration on this point. In the previously untreated patients, reversal of infectiousness occurred in 8 patients (35 per cent) who received isoniazid and thiocarbanidin, in 12 patients (55 per cent) who received isoniazid and PAS, in one patient (17 per cent) who was given streptomycin and thiocarbanidin, and in 2 patients (29 per cent) who received streptomycin and PAS. As expected, re-treatment patients showed a lower reversal rate. Tubercle bacilli

TABLE 3

PREVIOUSLY UNTREATED PATIENTS WITH BACILLI RESISTANT TO COMPANION DRUG

Regimen	Number of Patients with Resistant Bacilli	Number of Patients with Tubercle Bacilli in the Sputum at Onset of Study	Per Cent of Patients with Resistant Bacilli
Isoniazid-thiocarbanidin	9	23	39
Isoniazid-PAS	1	22	5
Streptomycin-thiocarbanidin	4	6	67
Streptomycin-PAS	3	7	43

disappeared from the sputum of 2 patients (13 per cent) who received isoniazid and thiocarbanidin, and of 2 patients (29 per cent) who were given isoniazid and PAS, but reversal of infectiousness failed to occur in any re-treatment patients who received streptomycin and thiocarbanidin or PAS.

Drug-susceptibility studies: The emergence of strains of tubercle bacilli resistant to the companion drug, streptomycin or isoniazid, was especially informative. Resistance to streptomycin or isoniazid was defined as equal growth in the control culture in streptomycin concentrations of 10 γ per ml., or any growth in streptomycin concentrations of 100 γ per ml.; and equal growth in the control culture in isoniazid concentrations of 1 γ per ml., or any growth in 5 γ of isoniazid per ml. In the previously untreated patients whose sputum was infectious at the start of the study, 9 of 23 patients (39 per cent) who received isoniazid–thiocarbanidin had strains resistant to isoniazid, and one of the 22 patients (5 per cent) who received isoniazid and PAS excreted a similar resistant strain (table 3). At the end of two months of therapy with isoniazid and thiocarbanidin, 3 of 14 previously untreated patients whose strains were tested were found to be excreting tubercle bacilli resistant to isoniazid, and none of 8 patients who received isoniazid–PAS had resistant bacilli at that time. In 6 of the previously untreated patients who received streptomycin–thiocarbanidin, strains of tubercle bacilli resistant to streptomycin appeared in 4 (67 per cent), while resistance was shown in the tubercle bacilli of 3 of the 7 patients (43 per cent) who were given streptomycin–PAS. On the completion of two months of therapy in previously untreated cases, 3 of the 5 patients who received streptomycin and thiocarbanidin whose strains were tested had streptomycin-resistant tubercle bacilli, but only one of the 5 patients who received streptomycin–PAS and whose strains were tested had resistant bacilli. Drug-resistance studies in the re-treatment group did not show the same marked difference in the appearance of resistance to the companion drug between all of the regimens containing thiocarbanidin and PAS as was shown in the previously untreated patients. Among those re-treatment patients who received isoniazid and thiocarbanidin, 5 of 16 patients (31 per cent) showed resistant bacilli, and 2 of 7 patients (29 per cent) who received isoniazid and PAS had resistant bacilli. Resistant bacilli appeared in 8 of the 13 re-treatment patients (62 per cent) who were given streptomycin and thiocarbanidin and who had infectious sputum at the onset of therapy, and only one of the 6 patients (17 per cent) who received streptomycin and PAS showed resistant bacilli (table 4). Less reliance can be placed on the drug-resistance studies in the re-treatment group because many of these patients were given both streptomycin and isoniazid previously, and some patients were included in the study whose bacilli initially were not completely susceptible to the isoniazid or streptomycin administered.

Roentgenograpic findings: No attempt was made to assess roentgenographic change other than an unfavorable change. In the previously untreated group, all of the patients who showed

TABLE 4

RE-TREATMENT PATIENTS WITH BACILLI RESISTANT TO COMPANION DRUG

Regimen	Number of Patients with Resistant Bacilli	Number of Patients with Tubercle Bacilli in the Sputum at Onset of Study	Per Cent of Patients with Resistant Bacilli
Isoniazid-thiocarbanidin	5	16	31
Isoniazid-PAS	2	7	29
Streptomycin-thiocarbanidin	8	13	62
Streptomycin-PAS	1	6	17

roentgenographic worsening were receiving thiocarbanidin with either isoniazid or streptomycin. None of the patients who received PAS with isoniazid or streptomycin had an unfavorable roentgenographic change. Unfavorable roentgenographic change occurred in 4 of 23 previously untreated patients (17 per cent) who received isoniazid–thiocarbanidin and in 2 of 7 patients (29 per cent) who were given streptomycin–thiocarbanidin (table 5). All of the previously untreated patients who showed roentgenographic worsening had bacilli resistant to the companion drug with the exception of one patient whose bacilli were only partially resistant to isoniazid. The re-treatment patients with unfavorable roentgenographic change did not show such striking correlation with a particular drug pairing. Four of 18 re-treatment patients (22 per cent) who received isoniazid–thiocarbanidin and 2 of 7 patients (29 per cent) who were given isoniazid–PAS showed roentgenographic worsening, while 5 of 15 re-treatment patients (33 per cent) who received streptomycin–thiocarbanidin and one of 6 patients (17 per cent) who were given streptomycin–PAS had unfavorable roentgenographic change (table 6).

One of the re-treatment patients who received isoniazid–PAS and showed roentgenographic worsening had only an increase in cavity size without increase in extent of disease. The other re-treatment patient who was given isoniazid–PAS and showed roentgenographic worsening had bacilli which were partially resistant to isoniazid at the onset of therapy. The re-treatment patients who received isoniazid with thiocarbanidin or PAS failed to show close correlation between roentgenographic worsening and isoniazid resistance but, among the 6 re-treatment patients who were given streptomycin and thiocarbanidin or PAS who had unfavorable roentgenographic change, resistance to streptomycin was present in 5.

TABLE 5

UNFAVORABLE ROENTGENOGRAPHIC CHANGE IN PREVIOUSLY UNTREATED PATIENTS

Regimen	Number of Patients with Roentgenographic Worsening	Number of Patients on Therapy	Per Cent of Patients with Roentgenographic Worsening
Isoniazid-thiocarbanidin	4	23	17
Isoniazid-PAS	0	24	0
Streptomycin-thiocarbanidin	2	7	29
Streptomycin-PAS	0	7	0

TABLE 6

UNFAVORABLE ROENTGENOGRAPHIC CHANGE IN RE-TREATMENT PATIENTS

Regimen	Number of Patients with Roentgenographic Worsening	Number of Patients on Therapy	Per Cent of Patients with Roentgenographic Worsening
Isoniazid-thiocarbanidin	4	18	22
Isoniazid-PAS	2	7	29
Streptomycin-thiocarbanidin	5	15	33
Streptomycin-PAS	1	6	17

Drug toxicity: There was no toxicity to isoniazid, streptomycin, or PAS other than the manifestations sometimes found, which consisted of gastrointestinal symptoms due to PAS, dizziness and rash due to streptomycin, and rash due to isoniazid. At the Veterans Administration–Armed Forces Chemotherapy Conference in February, 1959, the statement was made that there were no manifestations of toxicity to thiocarbanidin. Two patients were later reported to have developed fever presumably due to thiocarbanidin. One patient at the Veterans Administration Hospital in Memphis again had fever when thiocarbanidin alone was restarted. No report was made whether the other patient had another trial of therapy with thiocarbanidin; however, at the time he was febrile, he was also reported to have leukopenia. Thiocarbanidin was discontinued in both patients. One patient developed gastrointestinal distress due to thiocarbanidin requiring discontinuation of the drug but, as expected, there were more patients receiving PAS in whom it was necessary to stop the drug because of gastrointestinal symptoms. Another patient was reported to have a rash probably due to thiocarbanidin which was discontinued because of the emergence of tubercle bacilli resistant to streptomycin, the companion drug.

Drug concentrations in the blood: Measurements of thiocarbanidin in the blood done at the Memphis Veterans Administration Hospital in 11 patients who received 1 gm. of thiocarbanidin

daily in a single dose for one week showed concentrations two hours after receiving thiocarbanidin, varying from 0.2 γ of thiocarbanidin per ml. to 1.7 γ per ml., the average value being 0.9 γ per ml. The average concentration at six hours was 0.64 γ per ml., and the concentrations varied from 0.2 to 1.5 γ per ml. Dill and Glazko (3) found that the concentrations of drugs in two normal adult males receiving 1 gm. of thiocarbanidin daily in four divided doses reached maximal values of 1.2 γ per ml. and 1.4 γ per ml. After two normal males received 0.5 gm. of thiocarbanidin four times daily, the maximal concentration was 3.7 γ per ml. in one person, but was only 1.3 γ per ml. in the other subject. They determined the blood thiocarbanidin after one month of therapy in 5 patients who received 0.25 gm. of the drug twice daily and in 5 patients who received 0.5 gm. twice daily. The drug concentrations in the blood drawn two hours after receiving the morning dose of 0.5 gm. of thiocarbanidin showed a mean value of 0.63 γ per ml. Determinations made at the Memphis hospital on serum from 9 patients who had received 1 gm. of thiocarbanidin for a month showed considerable variation. Two hours after administration, the drug ranged from 0.2 γ per ml. to 2.0 γ per ml., and in one patient the value was 2.7 γ per ml. In 3 patients the concentration of drug in the blood was 2.0 γ per ml. The average two-hour drug concentration after therapy had been administered for one month was 1.3 γ per ml., and the average value at six hours was 1.2 γ per ml. Dill and Glazko (3) found appreciable quantities of drug in the blood two days after the last dose and continued appearance of the drug in the urine for several days after the last dose, indicating slow removal of the drug from the body.

Discussion

The small number of patients in the study, particularly in the streptomycin group, makes analysis of the results difficult, but certain impressions can be derived from the study. In the previously untreated patients, a greater difference was noticeable in the regimens containing thiocarbanidin or PAS than was the case in the re-treatment patients. The results in the former group were easier to interpret because they were not distorted by previous drug therapy and possible emerging bacterial resistance as in the case of some of the re-treatment patients. Reversal of infectiousness has been considered in all tuberculosis chemotherapy studies as an important indication of drug effectiveness. The previously untreated patients who received PAS with isoniazid or streptomycin showed a somewhat higher rate of reversal of infectiousness than did the patients who received thiocarbanidin with isoniazid or streptomycin. Of special significance is the fact that the previously untreated patients who showed unfavorable roentgenographic change had all received thiocarbanidin with either isoniazid or streptomycin; none of the patients who received PAS with streptomycin or isoniazid had roentgenographic worsening. The results in the re-treatment group are not entirely in agreement with this last finding. However, if the 2 patients with roentgenographic worsening who received isoniazid and PAS are discounted, because there was merely an increase in cavity size in one patient and in the other patient the tubercle bacilli showed some pretreatment resistance to isoniazid, the results would be compatible with those observed in the previously untreated patients.

The ability of a relatively weak drug such as thiocarbanidin or PAS to delay the emergence of bacterial resistance to a more effective drug (streptomycin or isoniazid) has also been considered as a measure of its usefulness. The results in the previously untreated patients indicated the superiority of PAS in delaying the appearance of bacterial resistance to the companion drug, and particularly to isoniazid. The emergence of resistance to the companion drug was followed by unfavorable roentgenographic change in 6 of the 13 previously untreated patients who were given thiocarbanidin with isoniazid or streptomycin and who showed resistant bacilli. By contrast, no unfavorable roentgenographic change occurred in the 4 previously untreated patients who received PAS and who had bacilli resistant to isoniazid or streptomycin.

There were no serious manifestations of toxicity to thiocarbanidin, but there were also no serious reactions to the other drugs in the study, although they are known to occur. In 3 of the 63 patients who received thiocarbanidin, it was necessary to discontinue the drug because of toxicity. Perhaps some serious toxic reactions would have been seen in the study if a larger group of patients had been studied for a long period of time.

All workers who have studied the absorption and urinary and fecal excretion of thiocarbanidin have commented on the poor absorption of the drug. It is possible that, if better absorption of the drug could be accomplished, the results in using the drug would be improved.

Summary

From the limited data available it appears that thiocarbanidin is of doubtful value as used in the present study. Reversal of infectiousness and less unfavorable roentgenographic changes occurred in a higher percentage of patients who received PAS than thiocarbanidin. Toxic manifestations which were not serious occurred in 3 patients who received thiocarbanidin, but required discontinuation of the drug.

Sumario

Resultados de la Terapéutica con Tiocarbanidina e Isoniacida o Estreptomicina

A juzgar por los limitados datos disponibles, parece que la tiocarbanidina posee valor dudoso según se usó en el estudio actual. Hubo viraje del esputo y menos desfavorables alteraciones radiográficas en un porcentaje mayor de los enfermos que recibieron PAS que con la tiocarbanidina. En 3 enfermos que recibieron tiocarbanidina se observaron manifestaciones tóxicas que no fueron graves, pero que exigieron la discontinuación del medicamento.

Resume

Résultats du traitement par la thiocarbanidine, l'isoniaside et la streptomycine

Si l'on en juge par le petit nombre d'observations actuelles, il semble que la thiocarbanidine est d'une valeur douteuse dans la presente étude. On obtient des recherches négatives pour le bacille tuberculeux dans l'expectoration et une transformation moins defavorable des images radiographiques chez un plus grand pourcentage de malades recevant du PAS que chez ceux traités par la thiocarbanidine. Des manifestations toxiques, qui ne furent pas graves apparurent chez trois malades recevant de la thiocarbanidine, ce qui necessita l'arrêt du traitement par ce médicament.

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MYCOBACTERIUM SALMONIPHILUM SP. NOV. FROM SALMONOID FISHES[1]

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(Received for publication May 27, 1959)

Introduction

The presence of mycobacteria in salmonoid fishes was first recorded by Earp, Ellis, and Ordal (1) in 1953. Acid-fast bacilli had previously been reported from other cold-blooded animals including fishes of fresh-water and marine origin; recent reviews have been presented by Vogel (2) and Parisot (3).

The initiation of an intensive search for bacteria of this group revealed their widespread distribution in salmonoid fish populations. Acid-fast bacilli have been demonstrated in livers and kidneys of salmon and trout in Washington, Oregon, and California, as well as from salmon taken in Alaskan waters. While the organisms are generally considered to be associated with fish of hatchery origin (4), they have also been found in young and adult fish from areas presumably supporting only wild populations. Wood and Ordal (4) indicate that the incidence of the tuberculosis-like infection is directly related to the length of time the fish are reared in the hatchery prior to release. This may result from a common hatchery practice of feeding viscera and carcasses from spawned-out adults to juvenile fish. Therefore, in many instances viable mycobacteria are present in the food of young fish.

The following species of Pacific salmon have been shown to harbor mycobacteria: *Oncorhynchus gorbuscha*, *O. kisutch*, *O. tshawytcha*, *O. keta*, and *O. nerka*. Acid-fast bacilli have also been detected in rainbow trout (*Salmo gairdneri gairdneri*), including the sea-run form, steelhead trout.

Attempts to culture acid-fast bacteria from diseased fish have not been uniformly successful despite the presence of enormous numbers of bacilli. A number of cultures have been obtained, however, and the following report is the first concerned with a study of the morphologic, cultural, and pathogenic characteristics of rapid-growing, acid-fast bacilli isolated from kidneys of salmonoid fishes.

Materials and Methods

The cultures used in this study were: *Mycobacterium sp.* (SC) isolated from an adult upstream migrant chinook salmon[2]; *Mycobacterium sp.* (OR) isolated from a two-year-old hatchery-held steelhead trout[3]; *Mycobacterium sp.* (CAR) isolated from a hatchery-held juvenile chinook salmon[4]; *Mycobacterium sp.* (CC) isolated from an adult upstream migrant steelhead trout[5]; *Mycobacterium fortuitum* ATCC 6842[6]; *Mycobacterium smegmatis*[6]; *Mycobacterium platypoecilus*[6]; *Mycobacterium phlei* NCTC 525[6]; *Mycobacterium ranae*[6]; *Mycobacterium marinum* ATCC 927[7]; *Mycobacterium piscium* ATCC 9819[7]; *Mycobacterium thamnopheus* ATCC 4445.[7]

The following media were used in the investigation: Czapek agar; heart infusion agar[8]; Sabouraud dextrose agar[8]; Littman oxgall agar[8]; dextrose agar[8] plus 5 per cent fresh Seitz-filtered bovine serum; litmus milk[8]; MR-VP broth[8]; 0.1 per cent tryptone broth[8]; Simmons citrate agar[8]; urea broth; nutrient broth plus 0.1 per cent potassium nitrate; blood agar base[8] plus 5 per cent human blood, 10 per cent Bacto gelatin,[8] dissolved in distilled water with pH adjusted to 7.0; medium 1 of Reed and Orr (5) for production of hydrogen sulfide; Dubos Tween®-albumin medium containing Tween 80 and serum albumin.[8] The basal inorganic nitrogen medium of Gordon and Smith (6) was employed for carbohydrate utilization studies, the carbon sources being sterilized by Seitz filtration.

The stock cultures were maintained on heart infusion agar slants and held at 19° or 25°C. Starter cultures were prepared by inoculation from the stock culture into a tube of Tween-albumin medium plus serum albumin and were incubated

[1] From the Bureau of Sport Fisheries and Wildlife, Western Fish Disease Laboratory, Seattle, Washington.

[2] Federal Fish Cultural Station, Spring Creek, Skamania County, Washington.

[3] Oregon State Fish Commission, Oakridge, Lane County, Oregon.

[4] Federal Fish Cultural Station, Carson, Skamania County, Washington.

[5] Washington State Game Department, Chambers Creek, Pierce County, Washington.

[6] Strains supplied through the kindness of Dr. Ruth E. Gordon, Rutgers University; and Dr. Ross Nigrelli, New York City Aquarium.

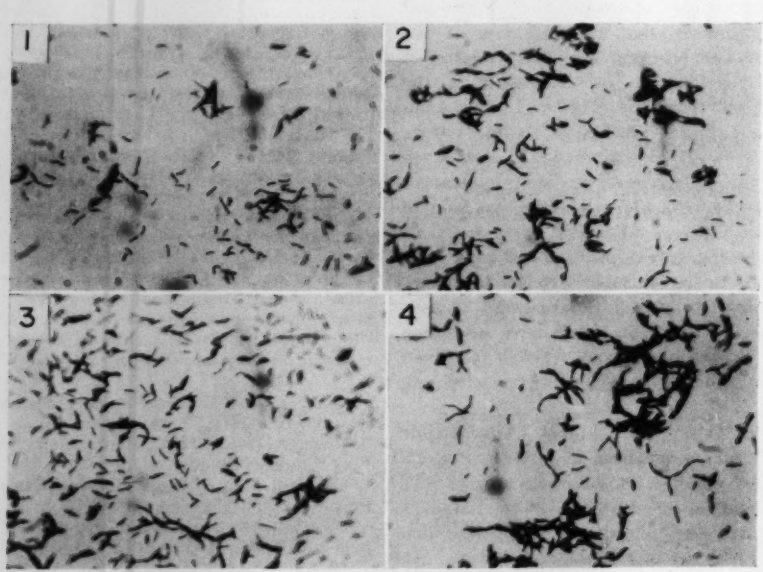
[7] Obtained from the American Type Culture Collection.

[8] Manufactured by Difco Laboratories.

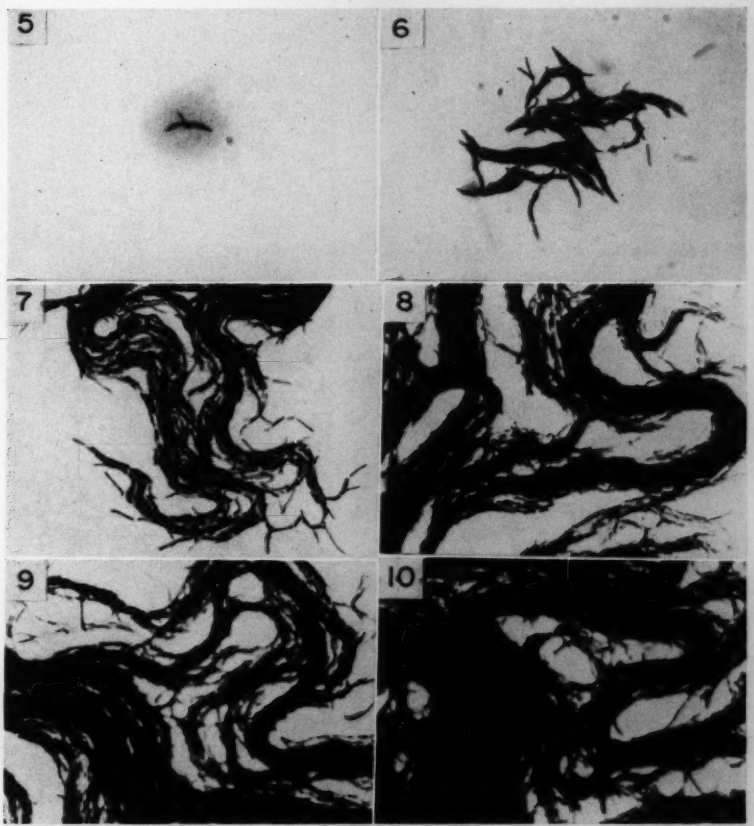
TABLE 1
ACID PRODUCTION FROM CARBOHYDRATES

Culture	Carbon Source														
	Dextrose	Mannose	Trehalose	Levulose	Mannitol	Xylose	Sorbitol	Sucrose	Rhamnose	Dulcitol	Arabinose	Galactose	Raffinose	Inositol	Lactose	Maltose
Group 1																
M. fortuitum	A	A	A	A	—	—	—	—	—	—	—	—	—	—	—	—
CC	A	A	A	A	A	—	—	—	—	—	—	—	—	—	—	—
OR	A	A	A	—	—	—	—	—	—	—	—	—	—	—	—	—
CAR	A	A	A	—	—	—	—	—	—	—	—	—	—	—	—	—
SC	A	A	A	—	—	—	—	—	—	—	—	—	—	—	—	—
Group 2																
M. smegmatis	A	A	A	A	A	A	A	—	A	A	A	A	—	A	—	—
M. ranae	A	A	A	A	A	A	A	—	A	—	A	A	—	A	—	—
M. phlei	A	A	A	A	A	A	A	—	—	—	A	A	A	—	—	—
Group 3																
M. piscium	A	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
M. marinum	A	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
M. platypoecilus	A	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Group 4																
M. thamnopheus	A	A	A	A	A	—	A	A	—	—	—	—	—	—	—	—

CC—strain isolated from steelhead trout in Chambers Creek hatchery.
OR—strain isolated from steelhead trout in Oak Ridge hatchery.
CAR—strain isolated from chinook salmon in Carson hatchery.
SC—strain isolated from chinook salmon in Spring Creek hatchery.



FIGS. 1–4. Photomicrographs of cultures isolated from salmonoid fishes:
FIG. 1. Strain from steelhead trout in Oak Ridge hatchery
FIG. 2. Strain isolated from chinook salmon in Spring Creek hatchery
FIG. 3. Strain isolated from chinook salmon in Carson hatchery
FIG. 4. Strain isolated from steelhead trout in Chambers Creek hatchery.



FIGS. 5–10. Photomicrographs of cultures isolated from chinook salmon in Carson hatchery at twelve-hour intervals to seventy-two hours.

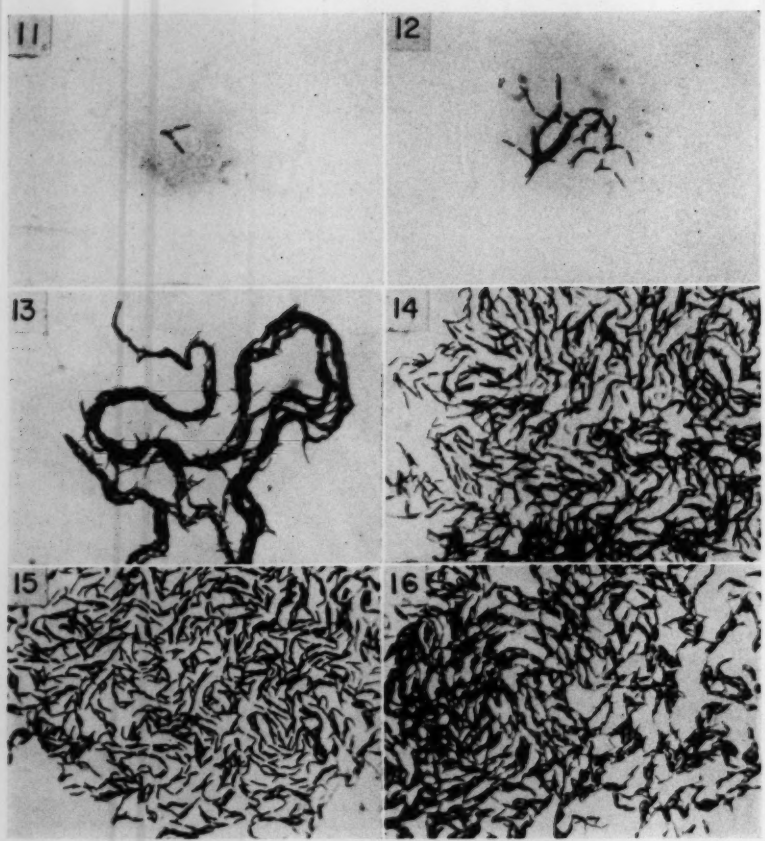
at 25°C. for four days. These cultures then served as the inoculum for morphologic, physiologic, and pathogenicity studies.

The acid-fastness was determined by staining smears obtained from cultures incubated for ten days at 19°C. and for four days at 25°C. The method of Ziehl-Neelsen was used.

The presence or absence of branching was determined as follows: clean glass slides were lightly scratched lengthwise with a diamond-point pencil. Several slides were placed in each of 4 wide-mouth bottles containing Tween-albumin medium plus serum albumin. Following sterilization each bottle was inoculated with one of the cultures under investigation. At twelve-hour intervals following inoculation, slides were removed, stained, and growth adhering to the area of the scratch was examined. Micro-slide cultures were prepared, using the technique of Knaysi (7) allowing the examination of growing cultures. Slide impression smears of microcolonies were made by punching out sterile discs of heart infusion agar medium and placing them in sterile Petri plates. A drop of dilute broth culture was placed on the disc and incubated at 25°C. At twelve-hour intervals a clean coverslip was carefully placed over a disc to make contact with the surface of the medium. The preparation was then placed in Bouin's fixative solution (8) for eight to twelve hours or until the disc had become completely impregnated with the fixing agent. The coverslip was carefully removed, washed in distilled water, and stained immediately. Permanent mounts were prepared with mounting fluid.

Neutral-red tests were conducted, using the methods of Dubos and Middlebrook (9) and Wayne (10).

Pathogenicity for mice was determined by injecting 0.5 ml. of four-day-old broth cultures into each of 2 young adult white mice (Swiss-



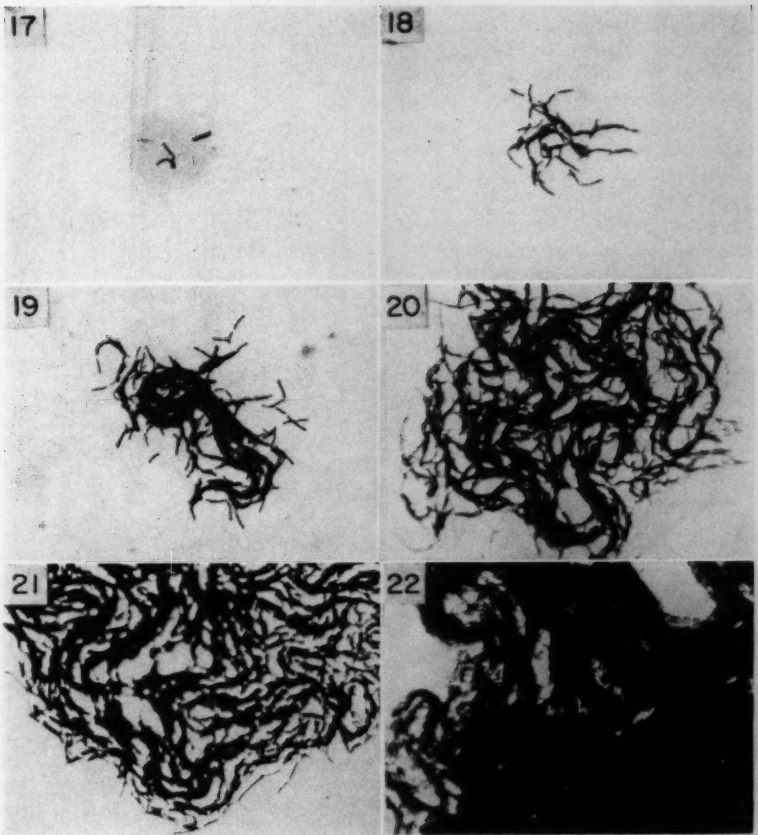
FIGS. 11–16. Photomicrographs of cultures isolated from chinook salmon in Spring Creek hatchery at twelve-hour intervals to seventy-two hours.

Webster), one receiving the organisms intraperitoneally, the other intravenously.

RESULTS

The 11 cultures were tested for carbohydrate utilization and could be divided into four distinct groups on the basis of acid production from the various carbon sources (table 1). *Group 1* included *M. fortuitum* and the 4 strains isolated from salmonoid fishes. *Group 2* was composed of the saphrophytic species and *M. ranae*. *Group 3* was made up of species previously recognized as fish pathogens, while *Group 4* consisted of a single pink-colored culture, *M. thamnopheus*.

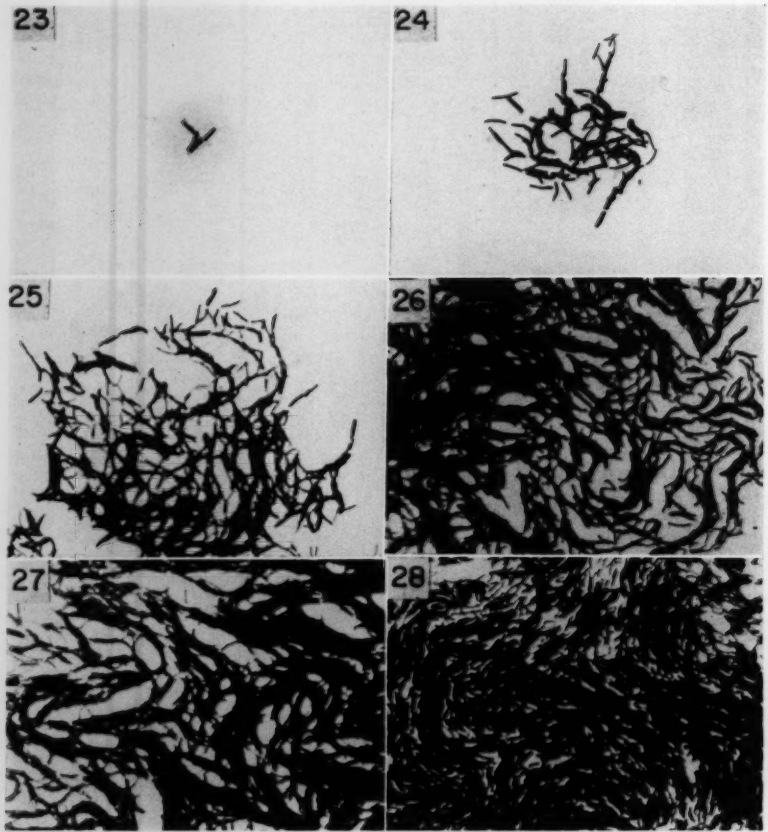
Group 1, off-white to pale yellow in color, was further differentiated from *Group 3* by virtue of the orange pigment produced by members of the latter group. Among the members of *Group 1*, the isolate from Chambers Creek was characterized by the production of a pale yellow pigmentation on medium containing serum. The formation of a water-soluble, purple pigment by strains SC and OR following extended incubation on Sabouraud dextrose agar served to distinguish these isolates from previously described mycobacterial species. Heavy inoculations of the organisms were planted on the culture medium in triplicate. Following incubation for two to three weeks at 19° and 25°C. in the absence of light, and at 25°C. in the presence of light, growth from isolates SC and OR turned a light purple color. Simultaneously, an amber-colored



FIGS. 17–22. Photomicrographs of cultures isolated from steelhead trout in Oak Ridge hatchery at twelve-hour intervals to seventy-two hours.

diffusible pigment appeared in the medium. The intensity of the coloration gradually increased and, approximately five to six weeks following inoculation, the colonial growth, as well as the medium, assumed a black-purple coloration. As might be expected, pigment production varied to some extent. The isolate from Oakridge temporarily lessened in its ability to form pigment. This became manifest as the colonial growth appeared light blue-gray and no diffusible pigment appeared in the medium. At the present time, however, maximal pigment production is evident. None of the other strains in the present investigation have ever been observed to produce this pigment.

Colonial morphology of the isolates from salmonoid fishes was similar to the description given for *M. fortuitum* by Gordon and Smith (11) although the culture from Chambers Creek tended to have a mucoid-like consistency. During the course of the investigation, a rough variant was selected from this strain. Isolates SC, OR, and CAR were identical in macroscopic appearance. On serum dextrose agar, incubated four days at 25°C., the colonies appeared light cream in color, round, convex, glistening, and smooth, ranging in size from punctiform to 1.5 to 2.0 mm. As the colonies aged and enlarged, the edges might spread and become flatter, the center appearing either heaped up or papilliform. Occasionally, faint radiating striations might be seen in older colonies. Following four days' incubation at 25°C. on Sabouraud dextrose agar and heart infusion agar, the appearance was similar to that



FIGS. 23–28. Photomicrographs of cultures isolated from steelhead trout in Chambers Creek hatchery at twelve-hour intervals to seventy-two hours.

seen on the above medium. Colonial size was reduced, the majority of colonies being punctiform. Colonies were light cream in color on heart infusion agar and off-white on Sabouraud dextrose agar. On these two media, particularly Sabouraud, the larger colonies did not exhibit the luster seen on serum dextrose agar. Occasionally, flat, dry, granular colonies with uneven, transparent edges were formed on all media.

Colonial morphology of the smooth culture from Chambers Creek differed in some respects from the other three isolates. The colonies were less convex, did not heap up in the center, or form papillae. They were mucoid-like in consistency, with coloration ranging from cream on Sabouraud dextrose agar to pale yellow on serum dextrose agar.

All four cultures were strongly acid fast when stained by the Ziehl-Neelsen method. Cells of isolates OR, SC, and CAR were morphologically similar when stained from four-day-old cultures taken from serum dextrose agar (figures 1–3). Straight or slightly curved forms were in evidence averaging about 1.5 to 2.0 microns by 0.25 to 0.35 microns. These cells stained evenly without evidence of beading with the exception of a few cells which contained dark, round or oval granules, usually terminal, but at times subterminal in location. Occasional filaments 4 to 6 microns in length were also seen; these appeared to be composed of several cells.

The majority of cells from CC strain were comparable in appearance and size to those described above; however, relatively long filaments were

TABLE 2
COMPARISON OF REACTIONS OF FOUR STRAINS OF MYCOBACTERIUM

Reactions	Strain			
	OR*	SC	CAR	CC
Citrate	+	+	+	+
Littman's oxgall agar	+	+	+	+
Nitrite from nitrate	−	−	−	+
Indol	−	−	−	−
Methyl red	−	−	−	−
Vosges-Proskauer	−	−	−	−
Gelatin liquefaction	−	−	−	−
Hydrolysis of starch	−	−	−	−
Hydrogen sulfide	−	−	−	−
Urease	−	−	−	−
Hemolysis	−	−	−	−
Litmus milk	Alkaline	Alkaline	Alkaline	Alkaline
Growth at:				
37°C	−	−	−	−
35°C	−	−	−	−
25°C	+	+	+	+
10°C	−	−	−	−

* See legends for table 1.

occasionally evident in preparations from the smooth phase (figure 4). These might be up to 10 microns in length and consisted of chains containing 6 or 7 discrete cells separated one from another by barely visible distances. Occasionally Y-shaped forms were present. These observations are in accord with Wykoff (12), who reported that branching forms of Mycobacterium are most commonly seen in the smooth phase.

Slide cultures were examined for the presence of branching. Microcolonies studied in the living state under low magnification did not show extensive branching. In fact, branching, if present at all, was extremely rudimentary. The difficulties encountered in making a determination of this kind were pointed out by McCarter and Hastings (13), who stated that apparent branching seen at lower magnifications may not be true branching. For this reason, slide impression smears, prepared at twelve-hour intervals from the microcolonies, were stained and examined, using an oil-immersion lens at a magnification of 1,000 ×. It becomes evident from the microphotographs (figures 5–28) that true branching does not occur, at least within the group of pigment-producing organisms under investigation. The microphotographs indicate that what may appear to be branching at lower magnifications is in reality an artifact. As the cells multiply, they show a tendency to orient themselves in parallel chains or packets composed of discrete curved or straight rods. Occasionally a cell will diverge at an angle from the longitudinal axis of the compound chain, giving the appearance of branching when examined at lower magnifications. Pseudobranching similar to this has been shown diagrammatically by Sguros (14) to occur in *Arthrobacter oxydans* of the family Corynebacteriacea.

Comparative tests of the biochemical activities and temperature requirements of the four cultures showed little difference (table 2). Biochemically, the CC strain differed from the others only in its ability to reduce nitrate to nitrite and in the utilization of levulose and mannitol. Temperature requirements of all cultures were identical.

White mice were not infected either by intravenous or intraperitoneal inoculation, and all cultures were neutral-red negative.

DISCUSSION

The four cultures isolated from salmonoid fishes appear to constitute a group of mycobacteria separate and distinct from any previously described species. While they resemble *M. fortuitum* Cruz most closely, their failure to grow at 37°C. and the production of a soluble pigment by two of the strains appear to preclude their identity with this species.

Schneidau and Shaffer (15), in a study of the cultural properties of strains of Nocardia, Streptomyces, and Mycobacterium, report that none of their strains of the latter organisms produced a soluble pigment. They state that association of such a pigment with an isolate would plead against its belonging to the genus Mycobacterium. However, Gordon and Mihm (16) have reported on strains, which they considered mycobacteria, that were capable of producing a diffusible melanin pigment. Also, Haag (17) reported a brown-black discoloration of culture media on which certain strains of acid-fast bacilli had been grown. He states that this pigment was very obvious in cultures of turtle and frog tuberculosis.

From this study, the isolates from Oakridge, Carson, and Spring Creek must be considered identical despite the fact that the latter culture has never been observed to produce pigment. This characteristic does vary among strains of Nocardia, and pigment-producing strains of *N. asteroides* have been reported by Schneidau and Shaffer (15).

If consideration is given to the possibility that the 3 cultures are members of the genus Nocardia, heed should be given to the statement of Waksman and Lechevalier (18), who report that the line of demarcation between Mycobacterium and the acid-fast Nocardia is recognized as difficult, if not impossible, and may rest upon the single characteristic of a branching mycelium which is considered specific for nocardias. Jensen (19) reports that the sometimes quite profuse branching in Mycobacterium and the slightly accentuated mycelial character of *Actinomyces farcineus* (*Nocardia farcineus*) tend to obscure the limit of separation. To add further confusion to the identification of these two groups, Waksman and Henrici (20) report that strains of actinomycetes may form a mycelium for a few transfers after isolation, and then appear forever after as acid-fast rods.

The existing taxonomic situation is summed up by Jensen (21), who states that, while the typical representatives of Mycobacterium and Nocardia are easy enough to recognize on the basis of morphology, staining reactions, and type of carbon metabolism, the genera are linked by boundary forms whose position is difficult or impossible to define. The distinctive features overlap, and each feature represents a spectrum that varies through many degrees.

Slide-culture preparations of the organisms under investigation, observed with the phase-contrast microscope at various periods of growth, have never yielded evidence of true branching. Impression smears, prepared at twelve-hour intervals over a seventy-two-hour period and examined with oil-immersion objectives at magnifications of 1,000 ×, have also failed to show the existence of true branching. Examination of growth adhering to scratched slide preparations has likewise yielded negative results.

Further support for the identification of the 4 cultures as Mycobacterium is gained through the strong qualities of acid-fastness exhibited by these organisms. Cells in smears prepared from month-old cultures on serum dextrose agar were approximately 100 per cent acid fast. The nocardias on the other hand were considered partially acid fast (22) with acid-fastness most apparent *in vivo* and not as marked in cultures.

McMillen and Kushner (23) used Littman oxgall agar as a means of differentiating the acid-fast nocardias from the Mycobacterium species. The nocardias used in their work failed to grow on this medium in contrast to the majority of strains of Mycobacterium which grow well on this medium, changing the color from blue to yellow. The 4 cultures reported herein from salmonoid fishes produced a heavy white moist growth and turned this medium yellow.

Another characteristic suggested by Schneidau and Shaffer (15), which appeared to be a useful criterion to separate strains of Mycobacterium from fragmenting Nocardia, is the inability of mycobacterial species to form a submerged mycelium. Of 49 strains of Nocardia studied by these authors, 48 produced a mycelium in the substrate, and the remaining strain was found to be quite unrelated to any of the "true" nocardias with which it had been compared. Eight of 10 strains of Mycobacterium failed to produce submerged mycelia. One of the 2 strains which formed a submerged mycelium was labeled *M. leprae* Duval 109, while the other strain was considered doubtful with regard to this characteristic. The formation of a submerged mycelium has never been observed in the 4 cultures under investigation.

While the 4 cultures exhibit a tendency toward cord formation, this characteristic is most pronounced in the Carson isolate (figures 5–11). As this culture was obtained from a population of fishes during an epizootic of a mycobacterial infection, a high degree of virulence may be anticipated for the strain. According to Dubos and Middlebrook (9), virulent forms of tubercle bacilli have a tendency to orient themselves in parallel in the direction of their long axis, often adhering to each other to give long serpentine "cords" while the markedly attenuated forms grow in a nonoriented manner.

On the basis of the information obtained in the present study, it is concluded that the organisms under discussion belong in the genus Mycobacterium. This distinction is based primarily on the observation that branching is completely lacking, or extremely rudimentary at most. The data provided in this study have revealed further that only one observed characteristic, the soluble pigment, may be considered as a property of the genus Nocardia. There is little to be found in the literature with regard to the appearance of most mycobacterial cultures on Sabouraud dextrose agar. While none of the other Mycobacterium species used in this study produced a soluble pigment on this medium, they obviously represent only a minute fraction of existing myco-

bacterial strains. A more extensive study of this property may indicate that production of soluble pigments by this group of organisms is not as rare as has been reported in the past.

The 4 cultures from Oakridge, Carson, Chambers Creek, and Spring Creek represent the first reported isolations of mycobacterial species from fishes of the family Salmonidae. In addition, a diffusible pigment is reported which is not present in currently recognized species of Mycobacterium. It is proposed that the isolates from Oakridge, Carson, and Spring Creek be designated *Mycobacterium salmoniphilum sp. nov.* in recognition of their affinity with salmonoid fishes. The isolate from Chambers Creek is designated *Mycobacterium sp. indeterminata* until such time as additional cultures can be isolated and examined.

Summary

Comparative morphologic, cultural, and pathogenic studies of rapid-growing mycobacteria recovered from kidneys of salmonoid fishes were conducted. The isolates were shown to differ from recognized species of Mycobacterium. In addition, the production of a purple, soluble pigment was reported for two of the strains. Three of the cultures were designated *Mycobacterium salmoniphilum sp. nov.* and one culture was designated *Mycobacterium sp. indeterminata.*

Sumario

El Mycobacterium Salmoniphilum sp. nov. Procedente de los Peces Salmonoides

Se llevaron a cabo estudios morfológicos, culturales y patógenos comparados de las micobacterias de rápido crecimiento obtenidas de los riñones de peces salmonoides. Se demostró que las aisladas discrepaban de las especies reconocidas del *Mycobacterium*. Además, se describió la producción de un pigmento purpurino soluble por dos de las cepas. Tres de los cultivos fueron denominados *Mycobacterium salmoniphilum sp. nov.* y un cultivo fué denominado *Mycobacterium sp. indeterminata.*

Resume

Mycobacterium salmoniphilum sp. nov. des poissons de la famille du saumon

On a étudié la morphologie comparée, la culture et le caractère pathogène de mycobactéries à developpement rapide, provenant des reins de ces poissons. Les bacteries isolées diffèrent des espèces connues de mycobacteries. En plus, on rapporte que deux de ces espèces produisent un pigment soluble violet. Trois de ces cultures furent nommées *Mycobacterium salmoniphilum sp. nov.* et une culture fut nommée *Mycobacterium species indeterminata.*

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Case Report

GAMMA GLOBULIN CONCENTRATIONS IN AMBULATORY PATIENTS WITH BRONCHIECTASIS[1]

A Survey with Report of One Case of Agammaglobulinemia Treated for Four Years

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(Received for publication July 16, 1959)

Introduction

Early in 1954, a year after the report by Janeway and associates (1) of a series of nine male children with absence of gamma globulin, two women on the medical wards of the Massachusetts General Hospital were found to have the same deficiency. Both women were admitted when critically ill; both had long-standing bronchiectasis; both died. These cases have been reported by Collins and Dudley (2).

This finding prompted the present writer to study the gamma globulin concentrations in the ambulatory hospital patients with bronchiectasis. It at once raised the hope that deficiency of gamma globulin might be the long-sought basic defect. Mallory (3) had explored the pathogenesis of bronchiectasis by a survey of 50 lobectomy specimens. His conclusion was that bronchial inflammation alone is rarely an effective factor but, in combination with atelectasis or pneumonitis, adequately accounts for most of the characteristic features of the disease. Attempts were made to find something which would explain the peculiar susceptibility of these patients to recurrent pneumonias and serious infections of the respiratory tract.

This study was undertaken to determine the extent of gamma globulin deficiency in the patients who came to the Pulmonary Clinic because of bronchiectasis. It has been in progress since 1954.[2] Meanwhile, Barrett and Volwiler (4) have reviewed the cases reported to November, 1956.

Plan of Study

An attempt was made to obtain blood samples from all patients with bronchiectasis now coming to the Pulmonary Clinic, as well as to bring back patients comprising the series reported in 1938 by Goodale (5) and in 1940 by Perry and King (6). These follow-up attempts were disappointing because of the difficulty of locating the patients after the lapse of so many years. Only 12 returned to the Clinic.

Janeway (1) had shown that demonstration of low or absent titer of isohemagglutinins would serve as a good screening procedure for absent or much diminished gamma globulin concentrations. Accordingly, blood from each patient was sent for blood grouping, Rh factor, and anti-A and B titers and for total protein, albumin, and globulin content and for gamma globulin determination. The latter was determined by paper chromatography.

Normal standards were: anti-A titer, 1:32 to 1:64, anti-B titer, 1:32 (occasionally 1:64); total globulin, 48 per cent of total protein; and gamma globulin, 17–18 per cent of total globulin in males, 20–22 per cent in females.

Samples were obtained from 52 patients with true bronchiectasis. All were ambulatory, some with mild, others with advanced, disease. Patients with bronchiectasis secondary to pulmonary fibrosis or the aging lung were not included in the study. Gamma globulin concentrations were obtained in 50 patients. One specimen was lost in a laboratory accident, and one was not obtained. In both of these patients (*Cases 21* and *49*) isohemagglutinin determinations were normal. Isohemagglutinins were obtained in 43 patients. Gamma globulin was present in the 9 on whom this information is lacking; in 2 instances (*Cases 27* and *37*), in increased amounts. In 2 instances (both group A), the anti-B titer was low, 1:4 (*Cases 15* and *50*). In no instance was a deficiency of gamma globulin found.

While this study was in progress a third woman with bronchiectasis, while ill in the hospital, was found to have a severe degree of

[1] From the Pulmonary Clinic of the Massachusetts General Hospital, Boston, Massachusetts.

[2] Funds for clinic visits and laboratory examinations were made available through the generosity of the Trustees of the Hospital.

TABLE 1

EXAMINATION OF BLOOD SAMPLES OBTAINED FROM 52 AMBULATORY PATIENTS WITH BRONCHIECTASIS

Case Number	Patient	Sex	Total Protein	Albumin	Globulin	Gamma Globulin	Blood Group	Anti-Isohemagglutinins		Remarks
			gm. per 100 ml.							
1	J. B.	M	6.8	4.4	2.4	Normal	O	A 1:128	B 1:128	
2	I. B.	F	7.3	4.9	2.4	Normal	B	A 1:16	—	
3	B. B.	F	7.0	4.2	2.8	Normal	A	Not obtained		
4	M. B.	F	7.09	4.43	2.65	Normal	O	Not obtained		
5	C. C.	F	7.2	4.8	2.4	Normal	O	A 1:16	B 1:32	*
6	G. C.	F	6.0	4.4	1.6	Normal	A	—	B 1:64	
7	A. C.	F	7.0	4.4	2.6	Normal	Not obtained	Not obtained		
8	G. C.	M	7.9	5.0	2.9	Normal	O	A 1:32	B 1:32	
9	R. C.	M	6.4	Not obtained		Normal	A	—	B 1:64	
10	J. C.	F	7.0	4.0	3.0	Normal	Not obtained	Not obtained		
11	C. C.	F	6.9	4.6	2.3	Normal	A	—	B 1:4	
12	C. C.	F	7.8	5.0	2.8	Normal	O	A 1:32	B 1:16	
13	W. D.	M	7.7	4.6	3.1	Normal	O	A 1:16	B 1:16	*†
14	J. E.	F	6.6	4.0	2.6	Normal	Not obtained	Not obtained		
15	M. G.	F	7.8	5.5	2.5	Normal	O	A 1:2	B 1:4	
16	M. G.	F	7.9	4.4	3.5	Normal	A	—	B 1:128	*†
17	T. G.	F	7.7	4.3	2.4	Normal	O	A 1:128	B 1:64	
18	A. G.	M	Not obtained			Normal	A	—	B 1:64	
19	L. G.	M	6.8	5.1	1.7	Normal	A	—	B 1:8	
20	J. H.	F	5.8	3.7	2.1	Normal	O	Not obtained		
21	G. H.	F	Lost	Lost	Lost	Lost	A	—	B 1:32	*
22	G. K.	F	8.0	4.9	3.1	Present	B	A 1:16	—	
23	B. L.	F	8.0	4.2	3.8	Increased	A	—	B 1:32	
24	L. L.	F	7.3	3.9	3.4	Increased	A	—	B 1:128	†
25	J. L.	F	7.3	4.2	3.9	Normal	O	A 1:256	B 1:16	
26	R. B.	M	7.8	4.6	3.2	Slightly increased	A–B	—	—	
27	B. M.	F	7.5	4.6	2.9	Increased	A	Not obtained		
28	F. M.	M	7.9	5.4	2.5	Normal	A	—	B 1:32	†
29	T. M.	M	7.5	4.3	3.2	Normal	A	—	B 1:8	
30	A. O.	F	Not obtained			Normal	O	A 1:32	B 1:32	
31	C. P.	F	7.4	4.9	2.5	Normal	O	A 1:4	B 1:32	*
32	N. P.	M	7.2	4.4	2.8	Normal	A	—	B 1:8	
33	L. P.	F	6.6			Normal	A	—	B 1:16	*†
34	D. R.	F	7.8	4.9	2.9	Normal	A	—	B 1:32	*†
35	J. R.	M	7.7	5.6	2.1	Normal	A	—	B 1:64	
36	D. R.	F	8.0	5.2	2.8	Normal	O	A 1:16	B 1:16	
37	E. S.	F	8.6	5.1	3.1	Markedly increased	O	Not obtained		
38	G. S.	M	7.5	5.1	2.4	Normal	O	A 1:4	B 1:32	
39	G. S.	M	6.8	4.7	2.1	Normal	O	A 1:32	B 1:16	
40	B. S.	F	6.9	5.4	1.5	Normal	A	—	B 1:32	*
41	P. M.	F	7.1	5.3	1.8	Normal	A	—	B 1:8	
42	T. V.	F	8.1	5.3	2.8	Present	B	A 1:32	—	
43	R. W.	F	Not obtained			Present	A–B	—	—	*†
44	M. W.	F	7.4	4.6	1.6	Present normal pattern	O	A 1:64	B 1:64	†
45	C. R.	F	8.0	5.0	3.0	Present	B	Not done		
46	J. F.	M	8.2	4.8	3.4	Present	A	—	B 1:16	
47	D. C.	M	7.1	4.3	2.8	Present	O	A 1:64	B 1:16	
48	A. R.	F	7.5	5.3	2.2	Slightly increased	O	A 1:32	B 1:16	
49	J. M.	M	7.5	5.1	2.4	Not obtained	O	A 1:64	B 1:16	
50	F. C.	M	8.0	5.1	2.9	Normal	A	—	B 1:4	
51	B. G.	F	8.0	5.2	2.8	Normal	A	—	B 1:64	
52	A. L.	F	6.7	4.3	2.4	Normal	A	—	B 1:32	

* Goodale case (5)
† Perry & King case (6)

hypogammaglobulinemia. This patient has been treated for four years. Her case is reported below:

Case Report

A 42-year-old woman was first seen in the outpatient department of the Massachusetts General Hospital in 1947 because of bronchitis. She stated that her chronic cough began at the age of fifteen following an attack of pneumonia. She never had whooping cough. Since the onset of cough she has had about twelve bouts of pneumonia. Her first admission to the wards of this hospital was in February, 1950, for study of her chronic lung disease. Her chief complaints were cough, fever, and wheezing. Skin tests with many allergens were negative. A roentgenogram of her chest in January, 1950, showed pneumonia of the left lower lobe. After the pneumonia cleared persistent atelectasis remained in the lobe and patchy density at the right apex. Sputum was negative for acid-fast bacilli twice. The tuberculin reaction was negative to 1:1,000 OT. The guinea pig inoculated with aspirated gastric contents died too early for diagnosis. The patient's weight in 1950 was 151 pounds. It was thought that she had bronchiectasis with repeated acute infections. She was discharged to the Pulmonary Clinic on prophylactic chemotherapy: 1 gm. of sulfamerazine daily. She has been followed in the Clinic since March, 1950. During the season 1950–1951 she had seven bad colds. Her weight dropped to 127 pounds. In August, 1951, the patient came to the emergency ward with pneumonitis of the right upper lobe and a temperature of 104.8°F.

In December, 1954, the patient had her second admission to the wards because of pneumonitis in the left lower lobe. This was several months after the first two cases of agammaglobulinemia had been seen in the hospital (3). Accordingly, studies of the patient's proteins were undertaken without delay. These and subsequent protein concentrations are set forth in table 2. The patient is Group A, Rh negative. On January 17, 1955, she was given I^{131}-labeled gamma globulin by Dr. David Gitlin at the Children's Hospital, Boston. The decay rate was approximately twenty-one days; this is normal or slightly longer than normal. The assumption was that she made gamma globulin too slowly to keep a normal concentration. As soon as these results were known, she was given her first dose, 30 ml. of gamma globulin, on March 14, 1955, and again returned to the outpatient clinic for follow-up observation. She reported that she felt very much better after the first dose.

Subsequent clinical course: Through the gener-

TABLE 2

Blood Values Obtained on Patient Reported

Patient M. P. (#57 88 88) Group A, Rh Negative

Date	Total Protein	Albumin	Globulin	Gamma Globulin	Isohemagglutinins Anti-B titer	Remarks
	gm. per 100 ml.					
12/12/54	6.4	5.9	0.5		1:32	Initial determination
12/17/54	6.05	5.05	1.0	Very low		Studied by electrophoresis
4/ 6/55	7.0	5.0	2.0			7 days after first 45 ml. of gamma globulin
5/11/55	6.5	5.5	1.0	7%		7 days after total 75 ml. of gamma globulin
7/20/55	6.6	5.4	1.2	5%		Continued on 15 ml. of gamma globulin every 14 days
1/25/56	6.4	5.5	0.9	"Low"		Patient very well Increased dose to 15 ml. of gamma globulin every 7 days
3/21/56	6.4	5.2	1.2			Continued dose of gamma globulin, 15 ml. every 7 days
6/13/57	6.8	5.3	1.5	"Low"	1:2	7 days after 67th consecutive weekly dose
10/10/57	5.1			8%	1:8	1 week after 83rd consecutive weekly dose
11/ 7/57	6.5			"Low"		5 weeks from previous injection
1/24/58	6.6	5.5	1.1	6%		16 weeks from previous injection
4/10/58	6.5	5.4	1.1	6%		6 months from previous injection
4/ 2/59	6.5	5.0	1.5	10%	1:16	Gamma globulin injections resumed 4/24/58 Patient is very well after 47 weekly doses of 15 ml. of gamma globulin.

TABLE 3
DETERMINATION OF PROTEINS AND ANTIBODIES IN PATIENT REPORTED

Blood specimen of October 10, 1957 (last injection of gamma globulin on October 3, 1957):		
Total serum protein	5.1 gm. per 100 ml. (low)	
Serum electrophoresis (filter paper):		
Albumins	65%	
α_1 globulins	8%	
α_2 globulins	7%	
β-globulins	13%	
γ-globulins	8% (low normal)	
Gamma globulins (immunochemical)	750 mg. per 100 ml. (low normal)	
Isohemagglutinins (Group A, Rh negative)	1:8 Anti-B (low normal)	
Influenza antibodies* (1 ml. of Asian strain influenza A vaccine September 12, 1957):		
Complement-fixation:		
vs. Influenza A	<1:8	
vs. Influenza B	<1:8	
Hemagglutinin inhibition vs. A strains		(no response)
A/PR8/1933	<1:12.5	
A/Fm$_1$/1947	<1:12.5	
A/Denver/1956	<1:12.5	
A/Jap 305/1957	<1:12.5	

* From the Massachusetts Department of Public Health Laboratories

osity of Dr. Charles Janeway of the Children's Hospital, the patient continued to receive 15 ml. of gamma globulin intramuscularly every two weeks. The concentrations of total and gamma globulins are presented in table 2. In the year 1955, the patient had "colds" with fever to 102° or 103°F. in March, April, May, June, August, and December.

After the finding of a total globulin of only 0.9 gm. per ml. in January, 1956, it was decided to decrease the interval between doses from two weeks to one. On March 21 the total globulin had risen to 1.2 gm., and the schedule of 15 ml. every seven days was continued until September 26, 1957. She had no severe or feverish cold between December, 1955, and her clinic visit on December 6, 1956. However, on December 27, 1956, she developed a respiratory infection with laryngitis, cough, and temperature of 102°F. This appeared to respond to 1 gm. of tetracycline (Achromycin®) daily for four days. On January 9, 1957, six days after the last previous injection of gamma globulin, her isohemagglutinins were 1:8 anti-B (low) compared to an initial normal level of 1:32. In 1957 she had respiratory infections of sufficient severity to cause increase in cough and sputum in February, May, and June. During this period she was receiving 15 drops of saturated solution of potassium iodide twice daily and, for much of the time, 2 gm. of sulfadiazine daily.

On September 12, 1957, the patient was given 1 ml. of influenza A vaccine, Asian strain. There was no antibody to this new strain of virus in the gamma globulin that had been injected, so that this provided an opportunity to see if she could form antibody. Dr. Janeway, who has most kindly advised us on the management of this patient, then suggested that gamma globulin injections be stopped for a time to see whether the patient really needed them. This was done after the eighty-third consecutive weekly injection on October 3, 1957. Blood drawn one week later was examined by Dr. Janeway whose report is presented in table 3.

At a clinic visit in January, 1958, sixteen weeks after the last injection, the patient stated that she thought she had made progress: she was able to go out in cold weather and to do all of her marketing; she had had no recurrent chills or fever and had regained about 15 pounds, which brought her weight to 140 pounds. She was being given a daily ration of 2 gm. of sulfadiazine. However, by April she seemed to be having an increased number of head and chest colds and was raising 30 ml. of green sputum daily. Blood drawn on April 10, 1958, six months after her last previous injection, showed a total globulin of 1.1 gm. per ml.; gamma globulin, 6 per cent. Injections were resumed one week later and she had 47 consecutive weekly doses of 15 ml. of gamma globulin. In October, 1958, she was sick at home with a temperature of 104°F., and in December she was still raising sputum, sometimes green. *Hemophilus influenzae* was cultured from the sputum. In January, 1959, the patient again had a lower respiratory infection, but without chills or significant fever.

The patient's current status in April, 1959, four years after the initiation of gamma globulin therapy, is as follows: Her weight is 132 pounds, she feels and looks very well. She has had no febrile episode for six months, although there have been severe colds and influenza-like illnesses in her community. She has had no pneumonia since December, 1954, prior to starting therapy. She no longer has "chills with every cold." She has a little cough and raises 5 to 10 ml. of white sputum daily. On examination there were many rales and a transient wheeze anteriorly on the right and in the axilla. The left lung is dry.

The roentgenogram shows no change from its appearance after the patient had pneumonia in 1950: atelectasis in the left lower lobe and patchy density at the right apex are still present. Her total globulin (seven days after injection) is 1.5 gm., with gamma globulin of 10 per cent. Anti-B titer is 1:16. This patient will probably require replacement therapy indefinitely. The dosage schedule will be determined by her clinical condition.

SUMMARY

While the suspected diagnosis of agammaglobulinemia is being made with increasing

frequency, it is not a common condition. Barrett and Volwiler (4) have stressed the need for correct diagnosis of the condition. The present writer has studied 52 patients with long-standing bronchiectasis and has failed to find a single instance of deficiency of gamma globulin in an ambulatory patient. One case of severe hypogammaglobulinemia in a woman with bronchiectasis is reported, with the results of four years of replacement therapy.

Sumario

Las Concentraciones de Globulina Gamma en los Enfermos Ambulantes con Bronquiectasia. Repaso con Presentación de Un Caso de Agammaglobulinemia Tratado por Cuatro Años

Aunque se hace con frecuencia cada vez mayor el diagnóstico de sospecha de agammaglobulinemia, no se trata de una dolencia común. Barrett y Volwiler (4) han recalcado la necesidad de un diagnóstico acertado de la enfermedad. El A. ha estudiado 52 enfermos con bronquiectasia de larga duración sin encontrar un solo caso de deficiencia de globulina gamma en un sujeto ambulante. Preséntase un caso de hipogammaglobulinemia grave en una mujer con bronquiectasia, con los resultados de cuatro años de terapéutica de substitución.

Resume

Taux de gamma-globulines chez des malades presentant des bronchiectasies, et traites de facon ambulatoire. Avec un rapport sur un cas d'agammaglobulinemie avec un traitement de quatre ans

Bien que le diagnostic d'agammaglobulinémie soit porté de plus en plus fréquemment, ce n'est pas une maladie commune. Barrett et Volwiler (4) ont insisté sur le besoin d'un diagnostic exact de cette maladie. L'auteur a etudié cinquante deux malades ayant un long passé de bronchiectasies, et n'a pas trouvé un seul cas de deficience en gamma globulines, chez ces malades ambulatoires. On a rapporté un cas d'hypogamma globulinémie grave chez une femme ayant des bronchiectasies et ayant depuis quatre ans un traitement de remplacement.

Acknowledgment

The writer is greatly indebted to Dr. Charles Janeway for his interest and help and for the generous supplies of gamma globulin provided for the patient whose case report is given.

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Notes

KANAMYCIN–TETRACYCLINE IN CHRONIC PULMONARY TUBERCULOSIS WITH COMMENTS ON THE AUDITORY TOXICITY OF KANAMYCIN[1]

There is a need to find a combination of antituberculous agents which does not include streptomycin, isoniazid, or PAS. Kanamycin and tetracycline represented one such possible combination. The problem with respect to kanamycin, however, was the fact that it closely resembled neomycin in both its biologic and chemical properties. Similarity of the antimicrobial spectrum and the development of cross resistance between kanamycin and neomycin had been observed.[2] The similarity in chemical structure of the two agents was also striking, both agents containing the diamine base 2-desoxystreptamine joined by glycoside linkages to diaminohexoses producing strongly basic compounds.[3, 4] Since parenterally administered neomycin was known to produce deafness as its chief toxic manifestation,[5-9] it was anticipated that deafness would be a problem with kanamycin as well and, indeed, this has proved to be the case. In spite of its known ototoxic potential, however, it seemed justifiable to try kanamycin in the present group of patients with far advanced tuberculosis and streptomycin- and isoniazid-resistant organisms because there seemed to be no other hope.

The present report is concerned with the occurrence of auditory toxicity in 12 patients with chronic tuberculosis who received kanamycin, 1.0 gm. per day intramuscularly, and tetracycline, 2.0 gm. per day by mouth. The original plan of investigation called for continuing all patients on the regimen for a period of six months. In none of the patients was it possible to continue kanamycin for the proposed period, primarily because of the development of auditory toxicity.

Abrupt appearance of auditory toxicity was the cause of the discontinuation of kanamycin in 11 patients. Subjective complaints were the rather sudden onset of tinnitus followed by complaints of hearing loss after receiving from 49 to 144 gm. of kanamycin. Six patients had pretreatment and serial audiograms made. All of these patients had had streptomycin in the past and all the pretreatment audiograms were abnormal, showing preexisting high-tone deafness (loss in the 2,000–8,000 cycles per second range). In these patients it was possible to document further perceptive loss in the high frequency (2,000–8,000 cps) range of 5 to 25 per cent.

Four patients without pretreatment audiograms who had previously received streptomycin had audiograms made at the time they complained of tinnitus and hearing loss. These audiograms also showed a marked falling off of perception of higher tones. Although some of this perceptive loss could have been present as a result of previous streptomycin treatment, it is believed that kanamycin was responsible for further loss.

Follow-up audiometric studies were performed in 7 patients. In no instance did high-tone perception improve three months after cessation of kanamycin. In 4 patients slight further loss occurred.

A representative series of audiometric curves is given in figure 1. The patient represented had previously received streptomycin, which probably accounted for the 40 per cent loss in the 3,000–8,000 cps range. Further loss of perception can be seen above 4,000 cps immediately after the cessation of 144 gm. of kanamycin, and three months later. In this patient there was some apparent improvement in low-tone perception, that is, below 512 cps.

Vestibular toxicity was less common than hearing loss, and developed in only 2 patients. Both of these patients were in the older age group, sixty-seven and seventy-three years, and both also had severe hearing loss.

Renal toxicity in the form of a rising blood urea nitrogen was the cause of discontinuation of kanamycin in only one patient, a 50-year-old woman with no evidence of pre-existing renal disease. Five additional patients showed transient

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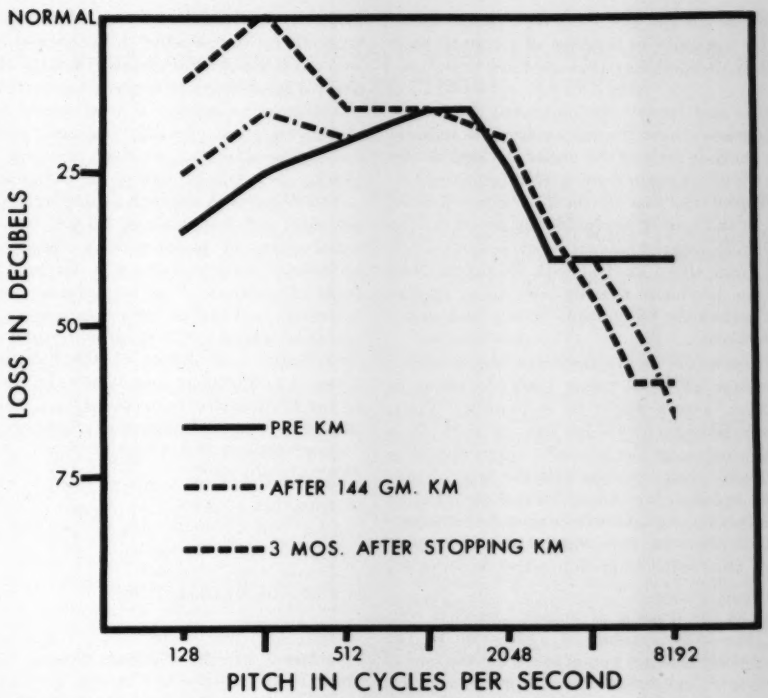

FIG. 1. Composite audiogram showing response to hearing for pure tones before, immediately after, and three months after treatment with kanamycin, 1.0 gm. per day.

rises in blood urea nitrogen (highest, 23 mg. per 100 ml.), but these rises were never sustained. The values returned to normal in spite of continued administration of kanamycin. Four patients showed diminution in twenty-four-hour creatinine clearances. All 12 patients in the study showed moderate intermittent cylindruria beginning between the third and seventh weeks, but this disappeared after cessation of kanamycin. Albuminuria, never more than a trace or one-plus, was infrequently and transiently seen in 4 of the 12 patients in the study.

There was no true reversal of infectiousness although 3 patients showed transient reductions in acid-fast bacilli on microscopy. Slight roentgenographic improvement occurred in one patient.

* * *

It is apparent that the limiting factor in the use of any regimen containing kanamycin in the 1.0-gm. per day dose range is the serious and irreversible auditory toxicity of kanamycin. This has been reported recently by others.[10-14] On the basis of the report of Schwartz,[14] in which kanamycin given in a dose of 1.0 gm. per day produced ototoxicity in twenty-nine of seventy-five patients, the Committee on Plans of the Eighteenth Veterans Administration–Armed Forces Conference on the Chemotherapy of Tuberculosis recommended abandoning the use of the daily 1.0-gm. dose of kanamycin. A new study was recommended employing lower doses such as 1.0 gm. twice weekly in re-treatment cases. Regimens containing kana-

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mycin in the 2.0-gm. per week dose range, as reported by Lattimer in a series of patients with renal tuberculosis,[15] may perhaps have more usefulness.

Hawkins and Lurie[16] demonstrated the site of action of neomycin in producing deafness in cats, namely, the hair cells of the organ of Corti of the cochlea. In physiologic studies these investigators demonstrated the loss of cochlear microphonics as well as the loss of nerve action potentials in the neomycin-treated cats. It seems reasonable to surmise from these studies with neomycin that kanamycin produces hearing loss in a similar manner, primarily by damage to the end organ (organ of Corti).

Some comments on the apparent improvement in perception of tones below 1,000 cps seems in order. Two factors might be responsible. First, this improvement could be due in part to a learning process, as the patients were unfamiliar at the time of initial testing with the type of pure test tone to listen for. An additional explanation is that kanamycin might reduce chronic nontuberculous infection in the middle ear and nasopharynx, thus improving conductive hearing for low tones.

So far as the hearing of human speech is concerned, the improvement on audiogram in the low frequencies cannot compensate for the loss of perception of high tones. Perception in the high-frequency range is important in the discrimination function. Unvoiced, sibilant, and consonantal sounds lie in the high-frequency range and, in human speech, are less intense than the vowels they accompany. Intelligibility of speech depends primarily on initial and final consonants.[17] This accounts for the patients' difficulty with conversation even though the hearing loss on the audiogram is limited to the higher frequencies.

Tetracycline appears to have added nothing to the effectiveness of kanamycin and, even in this low dosage (2.0 gm. per day), was not tolerated beyond six weeks by half of the patients.

In summary: A regimen of kanamycin, 1.0 gm. per day, and tetracycline, 2.0 gm. per day, in a small series of patients with chronic cavitary pulmonary tuberculosis, was an unsatisfactory form of therapy. The tetracycline was poorly tolerated, and half of the patients were unable to continue taking the drug for more than six weeks. The major toxic effect of the kanamycin was tinnitus and hearing loss, which developed in 11 of the 12 patients who received from 49 to 144 gm. of kanamycin. Audiometric studies revealed high-tone perceptive loss which persisted on retesting three months later.

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[16] HAWKINS, J. E., JR., AND LURIE, M. H.: Ann. Otol. Rhin. & Laryng., 1953, *62*, 1128.

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[18] The writer wishes to acknowledge the assistance of the following members of the staff of the University of Colorado School of Medicine: Drs. Paul Sutton and Donald Chen, Department of Surgery (ENT), for assisting in the evaluation of auditory toxicity; Dr. J. H. Holmes, Department of Laboratory Medicine, for making possible determinations of creatinine clearances; members of the resident staff, Drs. G. W. McDonough, I. Vinnik, K. Sussman, D. Armstrong, and J. Coe, for assistance in the care of the patients; Dr. Roger S. Mitchell for his encouragement and advice; and Miss Dorothy Nelson for preparing the manuscript.

THE RELATIVE EFFICACY OF CULTURE MEDIA FOR THE ISOLATION OF M. TUBERCULOSIS FROM CLINICAL MATERIAL[1]

Problems in the laboratory diagnosis of tuberculosis increased with the advent of mass roentgenographic survey programs for the general detection of early cases of tuberculosis and with the increasing use of antituberculous drugs even for nonhospitalized patients. More specimens containing smaller numbers of organisms are being submitted for examination to the laboratory. Drug-resistant bacilli as well as a variety of strains of mycobacteria are being cultured. Atypical acid-fast bacilli are observed more frequently than in the past. Therefore, periodic evaluation studies of the relative efficacy of different types of culture media for the isolation of these organisms are a continuing necessity.

The ideal culture medium for the isolation of tubercle bacilli from pathologic material has been described as having the following characteristics: "It should support rapid growth of small numbers of tubercle bacilli. The colony morphology should be such as to permit easy differentiation between pathogenic and nonpathogenic acid-fast bacilli. The medium should inhibit the growth of contaminating microorganisms. The medium should be simple to prepare from readily available ingredients."[2]

A variety of formulas of egg-potato medium as well as several semisynthetic types have been described for the cultivation of tubercle bacilli. Six of these media were selected for evaluation in this study.

The media evaluated and the formulas for each were as follows: Löwenstein-Jensen, (L-J),[3] American Trudeau Society (ATS),[4] Difco-Peizer TB (PTB),[5] Middlebrook 7H9 Agar A (7H9),[6] semisynthetic autoclavable (SSA),[7] and penicillin blood agar (PBA).[8]

The semisynthetic autoclavable medium[7] was prepared according to the published formula except that the sodium glutamate content was reduced from 40.0 to 4.0 gm., as recommended by the present writers.[9,10] The original formula for penicillin blood agar was modified and was as follows:

Agar (Difco)	1.5 gm.
Glycerin (CP)	1.0 ml.
Distilled water	74.0 ml.
Human blood bank blood (3 to 5 weeks old)	25.0 ml.
Penicillin (100 units per ml.)	

Because it was not possible to culture all specimens on all media at the same time, the procedure was as follows: Each medium was tested with not less than five thousand specimens per medium. Löwenstein-Jensen medium (used in this laboratory for approximately eight years) was the standard used throughout in determining the comparative efficacy of the other media.

A total of 27,271 specimens of sputum, gastric washings, et cetera, as received routinely in the Central Laboratory of the Florida State Board of Health, were used in the study. Most of the specimens arrived by mail and reached the laboratory two or more days after collection. Sputum specimens were submitted in sterile one-ounce bottles. Gastric washings and urine specimens were normally submitted in 100-ml. bottles containing approximately 35 ml. of a 10 per cent solution of trisodium phosphate.

The procedure employed routinely in this laboratory in the preparation of specimens for examination was used. To the specimens submitted without trisodium phosphate, an equal volume of a solution of 23 per cent trisodium phosphate was added. The solution was shaken for ten minutes in a modified Kahn shaker, and incubated overnight at 37°C. The next morning the specimens in their original bottles were centrifuged for twenty minutes at 2,800 to 3,000 r.p.m. in a No. 2 centrifuge equipped with Babcock-butterfat test centrifuge cups. Gastric washings and urine specimens were transferred to multiple one-ounce bottles for shaking and centrifugation. Sterile cotton-tipped swabs were used to transfer the sediment to the culture slants. The cultures were incubated at

[1] Supported in part by the U. S. Air Force under contract number AF (657)-26, School of Aviation Medicine, Randolph Air Force Base, Texas.

[2] MELVIN, I., *et al.*: Am. Rev. Tuberc., 1951, *63*, 459.

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[6] MORSE, W. C., *et al.*: Report No. 1, Fitzsimmons Hospital, Denver, Colorado.

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37°C. in ventilated wooden drawer-type boxes. They were examined for growth during the fourth, sixth, and eighth weeks of incubation. All cultures with colonies of sufficient size to be clearly identified were reported at the respective reading periods. Cultures with no growth after eight weeks were reported as negative. The same swabs used to inoculate the cultures were employed to prepare smears from the remaining sediment. The smears were stained by the Ziehl-Neelsen technique, examined microscopically, and stored for reference until the culture findings were reported.

For a number of years, it has been customary in this laboratory to inoculate two slants of Löwenstein-Jensen medium per specimen except for gastric washings, urine, and feces. For the latter, four slants per specimen are used. In the present study, one slant of the new medium being studied was added to the L-J slants as routinely used. The 27,271 specimens examined were divided into large series of not less than 5,000 each. For inoculation purposes, these were subdivided into smaller groups of 500. In these, the order of inoculation of the first and second slants (L-J and the test medium) were alternated. Thus, in the total series of specimens, the slants of L-J and the test medium which were being compared each received the first and the second inoculations an equal number of times. Therefore, the possible bias related to the order of inoculation was minimized. The *second* slant of L-J medium was always inoculated last. For gastric washings, urine, and feces, the number of slants was doubled but inoculated in the order indicated.

TABLE 1

RELATIVE EFFICACY OF LÖWENSTEIN-JENSEN MEDIUM AND PENICILLIN BLOOD AGAR MEDIUM FOR THE ISOLATION OF M. TUBERCULOSIS

	Number	Per Cent
Analysis of Specimens		
Total number cultured	5,849	100.0
Total positive for *M. tuberculosis*	523	8.9
Total negative for *M. tuberculosis*	5,176	88.5
Total unsatisfactory due to contamination	150	2.6
Distribution of Positive Findings		
L-J* positive, PBA positive	399	76.3
L-J positive, PBA negative	93	17.8
L-J negative, PBA positive	31	5.9
Total L-J positive	492	94.1
Total PBA positive	430	82.2

Comparison of Relative Rapidity of Growth

Incubation Time in Weeks	Initial Positive Findings on			
	L-J		PBA	
	Number	Per Cent	Number	Per Cent
4	319	64.8	253	58.8
6	127	25.8	119	27.7
8	46	9.4	58	13.5

Relative Frequency of Contamination

	Number	Per Cent
Total number of slants inoculated	12,264	
Number of L-J slants unsatisfactory due to contamination	261	2.1
Number of PBA slants unsatisfactory due to contamination	312	2.5

* L-J = Löwenstein-Jensen medium
PBA = penicillin blood agar

TABLE 2

RELATIVE EFFICACY OF LÖWENSTEIN-JENSEN AND AMERICAN TRUDEAU SOCIETY MEDIA FOR THE ISOLATION OF M. TUBERCULOSIS

	Number	Per Cent
Analysis of Specimens		
Total number cultured	6,161	100.0
Total positive for *M. tuberculosis*	275	4.4
Total negative for *M. tuberculosis*	5,783	93.9
Total unsatisfactory due to contamination	103	1.7
Distribution of Positive Findings		
L-J* positive, ATS positive	212	77.1
L-J positive, ATS negative	36	13.1
L-J negative, ATS positive	27	9.8
Total L-J positive	248	90.2
Total ATS positive	239	86.9

Comparison of Relative Rapidity of Growth

Incubation Time in Weeks	Initial Positive Findings on			
	L-J		PBA	
	Number	Per Cent	Number	Per Cent
4	148	59.7	144	60.3
6	51	20.6	40	16.7
8	49	19.7	55	23.0

Relative Frequency of Contamination

	Number	Per Cent
Total number of slants inoculated	12,798	
Number of L-J slants unsatisfactory due to contamination	198	1.6
Number of ATS slants unsatisfactory due to contamination	374	3.0

* L-J = Löwenstein-Jensen medium
ATS = American Trudeau Society medium

The relative efficacy of Löwenstein-Jensen medium and penicillin blood agar for the isolation of tubercle bacilli is shown in table 1. Löwenstein-Jensen medium detected 492 (94.1 per cent) of the positive specimens, compared with 430 (82.2 per cent) on PBA. On both media, 399 specimens (76.3 per cent) were positive for tubercle bacilli, while 93 (17.8 per cent) were positive on L-J and negative on PBA, and 31 (5.9 per cent) were positive on PBA and negative on L-J. This difference was found to be statistically significant. During the first four weeks of incubation, 64.8 per cent were clearly identified on Löwenstein-Jensen medium, compared with 58.8 per cent on penicillin blood agar medium. There was almost no difference in the rates of contamination between the two media, 2.1 per cent on the first and 2.5 per cent on the latter.

The comparison of the efficacy of Löwenstein-Jensen and American Trudeau Society media is given in table 2. Löwenstein-Jensen medium detected 248 (90.2 per cent) of the positive specimens, compared with 239 (86.9 per cent) on ATS medium. On both media, 212 (77.1 per cent) were positive for tubercle bacilli, while 36 (13.1 per cent) were positive on L-J medium and negative on ATS medium, and 27 (9.8 per cent) were positive on ATS and negative on L-J media. This difference was not found to be statistically significant. During the first four weeks of incubation,

TABLE 3

RELATIVE EFFICACY OF LÖWENSTEIN-JENSEN AND PEIZER TB MEDIUM (DIFCO) FOR THE ISOLATION OF M. TUBERCULOSIS

	Number	Per Cent
Analysis of Specimens		
Total number cultured	5,099	100.0
Total positive for *M. tuberculosis*	227	4.5
Total negative for *M. tuberculosis*	4,763	93.4
Total unsatisfactory due to contamination	109	2.1
Distribution of Positive Findings		
L-J* positive, PTB positive	147	64.7
L-J positive, PTB negative	66	29.1
L-J negative, PTB positive	14	6.2
Total L-J positive	213	93.8
Total PTB positive	161	70.9

Comparison of Relative Rapidity of Growth

Incubation Time in Weeks	Initial Positive Findings on			
	L-J		PTB	
	Number	Per Cent	Number	Per Cent
4	88	41.3	57	35.4
6	79	37.1	71	44.1
8	46	21.6	33	20.5

Relative Frequency of Contamination

	Number	Per Cent
Total number of slants inoculated	10,542	
Number of L-J slants unsatisfactory due to contamination	127	1.2
Number of PTB slants unsatisfactory due to contamination	229	2.1

* L-J = Löwenstein-Jensen medium
PTB = Peizer TB medium (Difco)

59.7 per cent of the positive specimens were clearly identified on L-J medium, compared with 60.3 per cent on ATS medium. There was a difference in contamination rates for the two media, 1.6 per cent on L-J and 3.0 per cent on ATS.

The relative efficacy of Löwenstein-Jensen medium and Difco-Peizer TB medium are shown in table 3. Löwenstein-Jensen medium detected 213 (93.8 per cent) of the positive specimens compared with 161 (70.9 per cent) on PTB medium. On both media, 147 (64.7 per cent) were positive for tubercle bacilli, while 66 (29.1 per cent) were positive on Löwenstein-Jensen medium and negative on PTB medium, and 14 (6.2 per cent) were positive on Peizer TB medium and negative on Löwenstein-Jensen medium. This difference was statistically significant. During the first four weeks of incubation, 41.3 per cent were identified on L-J medium, compared with 35.4 per cent on PTB medium. There was very little difference in contamination rates of the two media, 1.2 per cent on L-J and 2.1 per cent on PTB.

Löwenstein-Jensen and Middlebrook 7H9 Agar A (Difco) are compared in table 4. Löwenstein-Jensen medium detected 187 (95.9 per cent) of the positive specimens compared with 132 (67.7 per cent) on 7H9 Agar A. On both media, 124 (63.6 per cent) were positive for tubercle bacilli while 63 (32.3 per cent) were positive on L-J and negative on 7H9, and 8 (4.1 per cent) were positive on 7H9 Agar A medium and negative on L-J medium. This difference was statistically significant.

TABLE 4

RELATIVE EFFICACY OF LÖWENSTEIN-JENSEN AND MIDDLEBROOK 7H9 AGAR A MEDIA FOR THE ISOLATION OF M. TUBERCULOSIS

	Number	Per Cent
Analysis of Specimens		
Total number cultured	5,094	100.0
Total positive for *M. tuberculosis*	195	3.8
Total negative for *M. tuberculosis*	4,799	94.2
Total unsatisfactory due to contamination	100	2.0
Distribution of Positive Findings		
L-J* positive, 7H9 positive	124	63.6
L-J positive, 7H9 negative	63	32.3
L-J negative, 7H9 positive	8	4.1
Total L-J positive	187	95.9
Total 7H9 positive	132	67.7

Comparison of Relative Rapidity of Growth

Incubation Time in Weeks	Initial Positive Findings on			
	L-J		7H9	
	Number	Per Cent	Number	Per Cent
4	75	40.1	47	35.6
6	70	37.4	50	37.9
8	42	22.5	35	26.5

Relative Frequency of Contamination

	Number	Per Cent
Total number of slants inoculated	10,510	
Number of L-J slants unsatisfactory due to contamination	163	1.5
Number of 7H9 slants unsatisfactory due to contamination	137	1.3

* L-J = Löwenstein-Jensen medium
7H9 = Middlebrook 7H9 agar A medium

During the first four weeks of incubation, 40.1 per cent were identified on L-J medium, compared with 35.6 per cent on 7H9 Agar A. There was little difference in contamination rates between the two media, 1.5 per cent on L-J and 1.3 per cent on 7H9 Agar A.

The relative efficacy of Löwenstein-Jensen and the semisynthetic autoclavable medium is indicated in table 5. Löwenstein-Jensen detected 199 (97.1 per cent) of the positive specimens, compared with 97 (47.3 per cent) on SSA medium. On both media, 91 specimens (44.4 per cent) were positive for tubercle bacilli, while 108 (52.7 per cent) were positive on L-J medium and negative on SSA medium and 6 (2.9 per cent) were positive on SSA and negative on L-J media. This difference was statistically significant. During the first four weeks of incubation, 87 (43.7 per cent) were identified on L-J medium, compared with 20.6 per cent on SSA medium. There was little difference in contamination rates for the two media, 3.7 per cent on L-J and 3.2 per cent on SSA.

The number of positive cultures yielded by a medium was dependent in some degree on the order of inoculation. For comparison, there are the two Löwenstein-Jensen slants: one always inoculated as third and last, the other as the number one slant in one-half of the specimens and the number two slant in the remainder. Both L-J slants were positive for tubercle bacilli in 1,176 of the positive specimens. In 165 of the specimens, the L-J slant inoculated as first or second was posi-

TABLE 5

RELATIVE EFFICACY OF LÖWENSTEIN-JENSEN AND SEMISYNTHETIC AUTOCLAVABLE MEDIA FOR THE ISOLATION OF M. TUBERCULOSIS

	Number	Per Cent
Analysis of Specimens		
Total number cultured	5,068	100.0
Total positive for *M. tuberculosis*	205	4.1
Total negative for *M. tuberculosis*	4,634	91.4
Total unsatisfactory due to contamination	229	4.5
Distribution of Positive Findings		
L-J* positive, SSA positive	91	44.4
L-J positive, SSA negative	108	52.7
L-J negative, SSA positive	6	2.9
Total L-J positive	199	97.1
Total SSA positive	97	47.3

Comparison of Relative Rapidity of Growth

Incubation Time in Weeks	Initial Positive Findings on			
	L-J		SSA	
	Number	Per Cent	Number	Per Cent
4	87	43.7	20	20.6
6	54	27.1	25	25.8
8	58	29.2	52	53.6

Relative Frequency of Contamination

	Number	Per Cent
Total number of slants inoculated	10,416	
Number of L-J slants unsatisfactory due to contamination	392	3.7
Number of SSA slants unsatisfactory due to contamination	335	3.2

* L-J = Löwenstein-Jensen medium
SSA = Semisynthetic autoclavable medium

tive for tubercle bacilli while the other L-J was negative. Only 84 specimens were found positive on the second L-J slant when the first was negative. Considering all series, 1,425 specimens were found positive on either the first L-J slant or the test medium; 54 additional positive specimens were found only on the third and last L-J slant. Since the test media as a whole were less productive than L-J, it may be assumed that in a series of three L-J slants, the third would account for an even smaller number of added positive specimens. The proportion of all specimens found positive was 5.2 per cent, using two slants; and 5.4 per cent, using three. Thus, the inoculation of 27,271 additional slants revealed 54 additional positive findings, or one added positive per 505 additional slants inoculated.

* * *

Specimens submitted to public health laboratories vary in the amounts of material provided and in their condition on arrival. In too many, there is insufficient material for dependable findings. Some are relatively fresh on arrival, while others have deteriorated in transit. The technical problems are more difficult under these conditions than in situations in which an adequate volume of fresh specimens is collected and submitted.

Less than 4 per cent of the specimens cultured were gastric washings and/or urine specimens.

Each of these were inoculated to double the number of culture slants used for sputum. In tabulating the findings on gastric washings and urine specimens the two slants of each type were considered as one.

Almost twice as many specimens positive for tubercle bacilli were found in the penicillin blood agar series, compared with each of the other series. During this time, due to a temporary emergency, more than one thousand specimens with many positive cultures were submitted from one of the tuberculosis hospitals. All other series were the specimens normally received in the public health laboratories.

There is no agreement as to the desirable number of culture slants to be inoculated per specimen for the primary isolation of *M. tuberculosis*. Public health laboratories must consider economy in time and cost as well as sensitivity. In this study, it was found that the use of 3 culture slants as opposed to 2 increased the proportion of positive results from 5.2 to 5.4 per cent. This small increase was obtained by increasing by one-third the number of slants prepared and handled. Such an addition may be impracticable for laboratories required to process large numbers of specimens. The relative advantage of more slants per specimen versus encouraging more specimens per case must be weighed. In this laboratory, emphasis has been placed on the desirability of testing multiple specimens per patient. In addition, the sensitivity of the test has been increased by enlarging the surface. The surface which may be inoculated in a single one-ounce prescription bottle is equivalent to that in two tubes of medium customarily used in many laboratories. The procedure of using these two large slants per sputum specimen appears to be adequate and practicable.

Some difficulties were encountered in the preparation and use of the test media. Penicillin blood agar is relatively simple in composition but troublesome to prepare in large quantities. One of the major difficulties was the tendency of the blood to stratify in layers as the medium hardened, leaving the surface of the slant with the appearance of being overlaid with plain agar. Peizer TB medium and Middlebrook 7H9 Agar A were purchased from Difco and were easy to prepare. Semisynthetic autoclavable medium required more time and careful attention to detail. The presence of excessive surface moisture on this medium was a problem throughout all the weeks of incubation. It was quite easy to prepare the American Trudeau Society medium.

Colonies of tubercle bacilli growing on ATS and Peizer TB medium were comparable in appearance with those on L-J medium. They were usually dry, rough, breadcrumb-like, buff to cream-colored, and easy to recognize. On the other hand, colonies were much more difficult to identify on Middlebrook 7H9 Agar A and semisynthetic autoclavable media. Frequently, they were smooth and resembled yeast, particularly when the colonies were young. Those growing on PBA medium varied in color from pink to deep red and from gray to black, which made identification difficult for those unaccustomed to this medium.

In our hands, therefore, the Löwenstein-Jensen medium is slightly to substantially superior to the five possible alternates evaluated. Since the American Trudeau Society medium was almost as productive as L-J and was relatively easy to prepare, it would be nearly as desirable as the L-J for diagnostic use.

In summary: The relative efficacy has been compared of Löwenstein-Jensen medium, penicillin blood agar, American Trudeau Society medium, Difco Peizer TB medium, Middlebrook 7H9 Agar A medium, and a semisynthetic autoclavable medium for the isolation of *M. tuberculosis* from specimens submitted to a public health laboratory. Löwenstein-Jensen was the most sensitive of the media tested, although the difference in positive isolations on this and on the American Trudeau Society medium was not statistically significant. In this laboratory, the others were less effective and in some respects more difficult to use. A third slant increased only slightly the proportion of positive findings. The use of two slants with large areas for inoculation appears to be an acceptable technique, and this is practicable for laboratories handling large numbers of specimens.

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[11] The writers wish to thank the technical staff in the tuberculosis section of the laboratory for their assistance throughout this study.

THE C-REACTIVE PROTEIN IN CHILDHOOD TUBERCULOSIS[1]

The C-reactive protein, discovered by Tillett and Francis in 1930,[2] acquired its name as a result of their observation that it reacts with the C somatic polysaccharide of pneumococci to form a precipitate. First noted in the serum of patients acutely ill with various coccal infections or rheumatic fever, it was later found to be present in various other diseases which are characterized by inflammatory or necrotizing processes. Since the work of Anderson and McCarty in 1950,[3] it has been used widely as a measure of activity in acute rheumatic fever. The protein is not present in the serum of normal, healthy persons; therefore, any positive reaction may be considered abnormal.

Reports on the presence of C-reactive protein in the serum of patients with active tuberculosis have varied. Lofstrom[4] found that 17 of 23 patients with pulmonary tuberculosis in the early stage had positive C-reactive protein precipitin tests. Hedlund[5] reported that 13 of 19 patients with tuberculosis had positive tests. He states, however, that "every case with acute pulmonary tuberculosis is positive. The six negative (reactors) were those who had emerged from the active phase and were now in the healing stage." Roantree and Rantz[6] found C-reactive protein to be present in 3 of 7 early, active cases. On the other hand, Shetlar and associates[7] obtained positive reactions in 23 of 24 patients "with active tuberculosis." In a larger and more detailed study by Walsh and associates,[8] the patients were divided according to the extent of disease and activity. In the "active" group, they found that 9 of 15 patients with minimal disease were positive, 22 of 26 patients with moderately advanced disease were positive, and 32 of 34 patients with far advanced disease were positive. In the "inactive" group, only 6 of 35 were positive. In the recent Italian literature, Mescolini and Storniello,[9] reporting on 30 adult females, found that 88 per cent with exudative disease and 40 per cent with productive disease were positive; Carcassi and Pitzus[10] state that 4 patients with primary tuberculosis, 3 of 12 patients with post-primary disease, and 2 of 4 patients with tuberculous meningitis were positive.

The simple precipitin test used at present to detect the protein was devised by MacLeod and Avery,[11, 12] who showed that C-reactive protein was antigenic for rabbits and that specific antiserum could be prepared. The availability of commercial antiserum[13] and the use of a simple capillary tube method[14] make it a practical procedure which any clinical laboratory can perform.

The present report is a survey of 82 children[15] who were hospitalized with primary tuberculosis and/or its complications on the Children's Chest Service at Bellevue Hospital. Of these, 75 were children with active disease and 7 had inactive tuberculosis (calcified primary tuberculosis in 5, healed tuberculous meningitis in 2). In accordance with accepted procedure, the reactions were graded + to ++++++, depending upon the amount of precipitation. It was possible to repeat the test in 30 children, thus obtaining two or more serial values. Alpha-2 and gamma globulin determinations were also done, using the methods of Seibert[16] and Kunkel,[17] respectively. The alpha-2 is expressed in terms of Klett readings, the gamma globulin as Kunkel units (KU).

The upper limit of normal for the gamma globulin is considered to be 14 KU and for the alpha-2, 63. Total protein and albumin-globulin ratios in these patients were in the normal range. All determinations were done on the same sample of serum. No patient was accepted if another disease process

[1] This study was supported by research grants E-2482 and E-1281 from the Institute of Allergy and Infectious Diseases, National Institutes of Health, U. S. Public Health Service.

[2] TILLETT, W. S., AND FRANCIS, T., JR.: J. Exper. Med., 1930, *52*, 561.

[3] ANDERSON, H. C., AND MCCARTY, M.: Am. J. Med., 1950, *8*, 445.

[4] LOFSTROM, G.: Acta med. scandinav., 1942, *110*, 49.

[5] HEDLUND, P.: Acta med. scandinav., 1947, *128* (Supplement 196, p. 579).

[6] ROANTREE, R. J., AND RANTZ, L. A.: A.M.A. Arch. Int. Med., 1955, *96*, 674.

[7] SHETLAR, M. P., *et al.*: Proc. Soc. Exper. Biol. & Med., 1955, *88*, 107.

[8] WALSH, J. J., *et al.*: Am. Rev. Tuberc., 1956, *74*, 464.

[9] MESCOLINI, G., AND STORNIELLO, G.: Lotta contra tuberc., 1957, *27*, 1074.

[10] CARCASSI, V., AND PITZUS, F.: Minerva med., 1957, *48*, 2399.

[11] MACLEOD, C. M., AND AVERY, O. T.: J. Exper. Med., 1941, *73*, 183.

[12] MACLEOD, C. M., AND AVERY, O. T.: J. Exper. Med., 1941, *73*, 191.

[13] Manufactured by Schieffelin & Co., New York.

[14] SWIFT, H. F., *et al.*: J. Exper. Med., 1943, *78*, 127.

[15] The cases were consecutive except for a few instances in which serum was not obtainable.

[16] SEIBERT, F. B., *et al.*: Arch. Biochem., 1948, *18*, 279.

[17] KUNKEL, H. G.: Proc. Soc. Exper. Biol. & Med., 1947, *66*, 217.

TABLE 1

RESULTS OF INITIAL C-REACTIVE PROTEIN TESTS

Diagnosis	Number of Patients	Initial C-Reactive Protein*			
		Negative	+	++	+++
Primary tuberculosis	36	20	7	8	1
Tuberculous pleurisy	4	3	1	—	—
Tuberculous adenitis	3	1	1	1	—
Tuberculous bone disease	1	1	—	—	—
Protracted hematogenous dissemination	1	—	1	—	—
Miliary tuberculosis	2	—	2	—	—
Serous meningitis	1	—	1	—	—
Tuberculous meningitis	2	1	—	1	—
Subtotal (active cases)	50	26	13	10	1
Inactive tuberculosis					
Calcified primary tuberculosis	5	5	—	—	—
Healed tuberculous meningitis	2	1	1	—	—
Total	57	32	14	10	1

* Includes only those determinations taken during the first sixty days of hospitalization.

was present which could account for a positive C-reactive protein.

The results of the C-reactive protein precipitin test in children with primary tuberculosis or its complications are set forth in table 1. Only determinations done during the first two months of hospitalization are included. The children in the inactive tuberculosis group were readmissions to the hospital at the time when the study was done.

In their patients with rheumatic fever, Anderson and McCarty[3] noted: "All of the 45 patients studied . . . had moderate to large amounts of C-protein (+++ to ++++++) in the blood." By contrast, only one of the children in the present study had a precipitation of +++ and none had greater precipitation than that. Of the 57 patients included in table 1, 25 had positive C-reactive protein determinations and 32 negative. Excluding the inactive cases, 24 patients with early, active tuberculosis were positive, 26 negative.

It is important to note that the percentage of positive results was greater when the test was done early in the course of hospitalization (table 2). The children received isoniazid–PAS but, since treatment was not begun until the diagnosis had been established, the C-reactive protein determinations done during the first month of hospitalization were taken either before or shortly after the onset of therapy. Thus, as has been shown above, during the first two months of hospitalization, there was a fairly even division between positive and negative reactions (24 to 26 reactions). Of those taken after two months in the hospital, however, there were 24 negative reactions and only one positive.

Comparisons of the C-reactive protein with the gamma and alpha-2 globulins are made in tables tables 3 and 4. It was thought that these would be of interest for two reasons: First, these serum globulins have been found to be elevated consistently in tuberculosis.[7, 18-20] Second, in view of the evidence that the C-reactive protein belongs to the globulin fraction, it is of interest to determine whether any correlation can be demonstrated. (The C-reactive protein has been variously reported as being part of the alpha, beta, and gamma globulin fractions.)[5, 21, 22]

A comparison of C-reactive protein with gamma globulin is presented in table 3. Many of the patients who had elevated gamma globulin values had negative C-reactive protein determinations. Approximately half of these determinations were done early in the course of hospitalization. Nevertheless, relatively few samples (16 per cent) which showed negative C-reactive protein determinations showed *marked* elevation of the gamma globulin, i.e., more than 20 Kunkel units. There was better correlation when the C-reactive protein was positive; 62 per cent of the patients with positive C-reactive protein determinations had marked gamma globulin elevations and 14 per cent had slight to moderate elevations, making a total of 76 per cent correlation between positive C-reactive protein and elevated gamma globulin.

The data in table 4 indicate that there is good correlation between the C-reactive protein and alpha-2 globulin. Of those who had negative C-reactive protein determinations, 70 per cent had normal alpha-2 values; of those with positive C-reactive protein determinations, 77 per cent had elevated alpha-2 values. It has been established that the gamma globulin is elevated in minimal tuberculous disease, whereas the alpha-2 becomes elevated only when disease is moderately to far advanced, with tissue destruction. The

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[18] SEIBERT, F. B., *et al.*: J. Clin. Invest., 1947, *26*, 90.

[19] VOLK, B. W., *et al.*: Am. Rev. Tuberc., 1953, *67*, 299.

[20] ZITRIN, C. M., *et al.*: A.M.A.J. Dis. Child. (in press).

[21] WOOD, H. F., *et al.*: J. Exper. Med., 1954, *100*, 71.

[22] HEDLUND, P.: Scandinav. J. Clin. & Lab. Invest., 1955, *7*, 99.

TABLE 2

INITIAL C-REACTIVE PROTEIN IN RELATION TO LENGTH OF HOSPITALIZATION

Diagnosis	Number of Patients	1–14 days		15–30 days		31–60 days		61–90 days		over 90 days	
		Negative	Positive	Negative	Positive	Negative	Positive	Negative	Positive	Negative	Positive
Primary tuberculosis	55	6	5	4	5	10	6	13	—	5	1
Tuberculous pleurisy	8	1	1*	—	—	2	—	2	—	2	—
Tuberculous adenitis	3	1	—	—	1	—	1	—	—	—	—
Tuberculous bone disease	1	1	—	—	—	—	—	—	—	—	—
Primary tuberculosis with cavitation	1	—	—	—	—	—	—	1	—	—	—
Protracted hematogenous dissemination	1	—	1	—	—	—	—	—	—	—	—
Miliary tuberculosis	3	—	—	—	—	—	2	—	—	1	—
Serous meningitis	1	—	—	—	—	—	1	—	—	—	—
Tuberculous meningitis	2	—	1	1	—	—	—	—	—	—	—
Subtotal (active cases)	75	9	8	5	6	12	10	16	0	8	1
Inactive tuberculosis											
Calcified primary tuberculosis	5	5	—	—	—	—	—	—	—	—	—
Healed tuberculous meningitis	2	1	1	—	—	—	—	—	—	—	—
Total	82	15	9	5	6	12	10	16	0	8	1

* Treated with cortisone.

TABLE 3

COMPARISON OF C-REACTIVE PROTEIN WITH GAMMA GLOBULIN*

Diagnosis	Number of Patients	CRP† Negative	Gamma Globulin			CRP Positive	Gamma Globulin		
			14 or Less	15–20	20 or More		14 or Less	15–20	20 or More
Primary tuberculosis	79	49	21	20	8	30	7	5	18
Tuberculous pleurisy	14	12	3	9	0	2	2	0	0
Tuberculous adenitis	5	2	0	2	0	3	0	1	2
Tuberculous bone disease	1	1	1	0	0	0	0	0	0
Primary with cavitation	2	2	0	1	1	0	0	0	0
Protracted hematogenous dissemination	2	1	0	0	1	1	0	0	1
Miliary tuberculosis	3	1	1	0	0	2	1	0	1
Serous meningitis	1	0	0	0	0	1	0	0	1
Tuberculous meningitis	4	1	0	0	1	3	0	0	3
Total	111	69	26	32	11	42	10	6	26

* This represents total determinations; therefore, two or more were done on some patients.
† CRP—C-reactive protein.

good correlation between C-reactive protein and alpha-2 suggests that greater disease is necessary to demonstrate the presence of this abnormal protein than to cause an elevation of the gamma globulin.

The results of the C-reactive protein determination in the infants less than one year of age at the time of the study (age range of four to twelve months) are listed in table 5. The information was believed to be pertinent, since infants sometimes react in a manner different from that of older children and adults. Although the number of patients is small, the table indicates that they follow the same pattern as the older children.

Thirty children had serial determinations of the C-reactive protein. Most of those who were

TABLE 4

COMPARISON OF C-REACTIVE PROTEIN WITH ALPHA-2 GLOBULIN*

Diagnosis	Number of Patients	CRP† Negative	Alpha-2 Globulin		CRP positive	Alpha-2 Globulin	
			63 or Less	64 or More		63 or Less	64 or More
Primary tuberculosis	70	44	30	14	26	5	21
Tuberculous pleurisy	12	9	7	2	3	1	2
Tuberculous adenitis	5	2	2	0	3	1	2
Tuberculous bone disease	1	1	0	1	0	0	0
Primary with cavitation	2	2	1	1	0	0	0
Protracted hematogenous dissemination	1	0	0	0	1	1	0
Miliary tuberculosis	3	1	1	0	2	1	1
Serous meningitis	1	0	0	0	1	0	1
Tuberculous meningitis	4	1	1	0	3	0	3
Total	99	60	42	18	39	9	30

* This represents total determinations; therefore, two or more were done on some patients. However, since there was insufficient serum in some instances, it will be noted that the total number for gamma globulin is greater than for alpha-2.

† CRP—C-reactive protein.

TABLE 5

INITIAL C-REACTIVE PROTEIN IN INFANTS ONE YEAR OF AGE OR LESS

Diagnosis	Number of Patients	1–14 days		15–30 days		31–60 days		61–90 days		over 90 days	
		Negative	Positive	Negative	Positive	Negative	Positive	Negative	Positive	Negative	Positive
Primary tuberculosis	9	2	1	1	2			2		1	
Tuberculous adenitis	2	1			1						
Primary tuberculosis with cavitation	1							1			
Protracted hematogenous dissemination	1		1								
Tuberculous meningitis	1			1							
Total	14	3	2	2	3	0	0	3	0	1	0

initially positive became negative promptly. All received antituberculous therapy and recovered uneventfully. However, 5 of the children still had positive C-reactive protein determinations three to five months after admission. Three of these had primary tuberculosis (one complicated by varicella during the first month); one with pleural effusion took longer than usual to recover; and one had tuberculous meningitis following measles.

The results of this study in children, as well as the reports in the literature, suggest that a positive C-reactive protein is of greatest diagnostic value in tuberculosis in those equivocal cases in which it is difficult to determine whether the disease is active. In such instances, a positive test indicates the presence of activity. As a prognostic aid, its value lies in serial determinations. Given an initially positive test, conversion to negative appears to be a good prognostic sign.

In summary: In a group of 82 children with primary tuberculosis or its complications, the C-reactive protein was found to be positive in 24 of 50 patients with early, active disease, negative in 24 of 25 treated children who had been in the

hospital more than two months, and negative in 6 of 7 patients with inactive disease (the positive test occurring in a child with healed tuberculous meningitis). The gamma globulin was abnormal more often than the C-reactive protein test. There was good correlation, however, between the latter and the alpha-2 globulin. Although a negative test does not preclude activity, a positive test is good evidence for the presence of active disease and is most helpful in equivocal cases as confirming evidence of active tuberculosis. When the C-reactive protein is positive early in the disease, conversion to negative with serial determinations appears to be a good prognostic sign.

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SARCOIDOSIS[1]

Experience at a Naval Hospital

A diagnosis of sarcoidosis should be considered for every patient presenting with hilar adenopathy. If there is associated infiltration in the parenchyma of the lung, the case for sarcoidosis is strengthened. The rather startling pathology revealed in a roentgenogram of the chest often prompts the roentgenologist to call the referring physician for clinical data on this seriously ill individual, only to be greeted by the statement that the patient has gone on liberty or is out on a work detail. Such is frequently the experience on the chest service of a naval hospital where a majority of these patients are asymptomatic, admission being prompted by abnormal shadows in a roentgenogram taken as part of a routine examination.

It is the purpose of this paper to report the writers' experience with sarcoidosis, emphasizing the usual patterns seen. No attempt will be made to enter into the arguments regarding etiology or to review the literature. This has been done admirably in footnotes[2] through[6]. Two extensive, well-documented series have been reported recently.[7, 8]

The records of 59 cases examined consecutively, in which a diagnosis of sarcoidosis was made between 1947 and 1958, were reviewed. The diagnosis in each case was based upon excised tissue examined histologically and a clinical picture compatible with this disease. Of interest is the fact that 51 per cent of the patients were admitted to the hospital because of abnormal shadows observed in a roentgenogram obtained as part of a routine examination; 12 per cent were discovered by roentgenograms taken at the time of admission for other conditions; and 37 per cent came to the attention of a physician because of minor symptoms attributable to sarcoidosis.

Age, race, and state of birth: The age range was eighteen to forty-three years at the time of diagnosis. The median age was twenty-two years, slightly lower in the Negroes (twenty years) than in the white patients (twenty-three years). Thirty-six per cent were Negroes, an incidence ten times that of other races when corrected for the ratio of Negroes to other races in the Navy and Marine Corps. Only 2 patients were female. As observed by most other investigators,[9, 10] a majority (31 of this group) were born in the southeastern part of the United States.

Symptoms: Cough and malaise were mentioned most often. The cough was dry or only slightly productive. Hemoptysis was not recorded in a single case. The malaise was usually described as a generalized weakness or easy fatigability. Dyspnea was mild (exertional only) and was moderate in only one case of extensive parenchymal involvement of the lungs. Weight loss was observed in 12 of the patients and was greater than 10 pounds in 5 patients. Chest pain, when it occurred, was described as a vague tightness.

Signs: Certainly a majority of the patients appeared to be in excellent health. Although 59 per cent showed lymphadenopathy (generalized in 24 per cent), this was not marked and was found only upon careful examination of the peripheral lymph nodes. The cervical and axillary nodes were most frequently involved. Abnormal physical signs detected on examination of the chest were recorded in 27 per cent of the patients, but these, too, were insignificant and by no means diagnostic. Fever was of low grade, persisting for a few days or weeks and never going above 100°F. unless other unrelated disease was present.

Laboratory findings: Leukopenia with a leukocyte count of less than 5,000 per cu. mm. was observed in 15 per cent of the patients. Eosinophilia of 4 per cent or more was observed in a third of the patients. Hemoglobin determinations fell within the normal or low normal range. One-half of the patients showed elevations of the sedimentation rate. A total serum protein of greater than 8 gm. per 100 ml. was found in 15 per cent and a serum globulin of more than 3.5 gm. per 100 ml. in 39 per cent of the patients. Electrophoretic analysis of serum proteins obtained in 7 patients showed no consistent pattern. Serum calcium levels were only slightly elevated in 2 of 32 patients, neither

[1] From the Medical Service, United States Naval Hospital, Oakland, California. The opinions or assertions contained herein are those of the writers and are not to be construed as official or necessarily as reflecting the views of the Medical Department of the Navy or the Naval Service at large.

[2] FREIMAN, D. G.: New England J. Med., 1948, *239*, 664; 709; 743.

[3] LONGCOPE, W. T., AND FREIMAN, D. G.: Medicine, 1952, *31*, 1.

[4] MICHAEL, M., JR.: Am. J. M. Sc., 1958, *235*, 148.

[5] SILTZBACH, L. E.: Postgrad. M. J., 1958, *34*, 254.

[6] DUNNER, E., *et al.*: South. M. J., 1957, *50*, 1141.

[7] JAMES, D. G.: Brit. M. J., 1956, *2*, 900.

[8] ISRAEL, H. L., AND SONES, M.: A.M.A. Arch. Int. Med., 1958, *102*, 766.

[9] MICHAEL, M., JR., *et al.*: Am. Rev. Tuberc., 1950, *62*, 403.

[10] CUMMINGS, M. M., *et al.*: Postgrad. Med., 1956, *19*, 437.

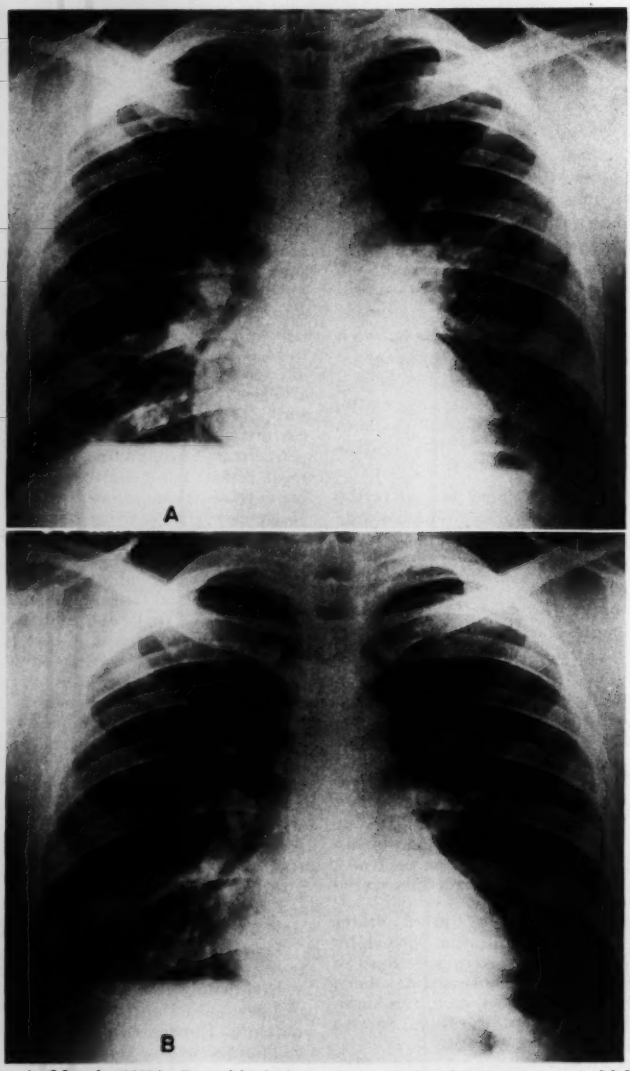

FIG. 1 (C. S.). *A* (March, 1957). Sarcoidosis in an asymptomatic twenty-year-old Negro male, discovered by routine roentgenogram of the chest; diagnosis confirmed by right scalene node biopsy. Note diffuse parenchymal infiltration and bilateral hilar adenopathy.

B (August, 1957). Same case five months later. Adrenal cortical therapy was given for seven weeks after the diagnosis was established. In view of negative reaction to first- and second-strength PPD, no antituberculous drugs were given. Note moderate clearing of parenchymal disease and decrease in size of adenopathy.

of whom showed bone lesions or renal damage. Pulmonary function tests were of little help in assessing the extent of the involvement.

Tuberculin skin tests were positive to intermediate-strength (0.0001 mg.) purified protein derivative (PPD) in 11 per cent. This compares favorably with the 9.7 per cent reactor rate to the same dose of PPD found in Navy recruits.[11] An

[11] CANADA, R. O., AND BABIONE, R. W.: Am. Rev. Tuberc., 1950, *62*, 518.

TABLE 1

ROENTGENOGRAPHIC MANIFESTATIONS OF MEDIASTINAL ADENOPATHY AND PARENCHYMAL INFILTRATION

Manifestations	Patients	
	Number	Per Cent
Parenchymal infiltration only	6	10
Mediastinal adenopathy only	26	44
Mediastinal adenopathy and parenchymal infiltration	26	44
Normal chest roentgenograms	1	2
Bone lesions (46 cases in which survey included hands and feet)	2	4

TABLE 2

DIAGNOSIS BY BIOPSY

Site of Biopsy	Patients	
	Number	Per Cent Positive
Scalene nodes	50	72
Peripheral nodes	16	75
Lung	9	100
Mediastinal nodes	5	100
Liver	1	100

equal percentage of subjects failed to react to intermediate-strength PPD but gave positive reactions when the second-strength (0.005 mg.) was applied. Ten patients with negative tests to intermediate-strength PPD did not have second-strength tests.

Roentgenographic manifestations: As mentioned at the outset, the roentgenographic manifestations are the most impressive features of this disease. Only one patient failed to show evidence of involvement of the lungs or mediastinal nodes. As indicated by figure 1, the parenchymal and hilar involvement could be extensive with only minimal symptoms. The incidence of mediastinal adenopathy and parenchymal infiltration observed is given in table 1. Cystic bone lesions in the hands or feet were observed in only 2 of the 46 cases surveyed.

Diagnosis: Removal of lymph nodes contained in the fat pad beneath the scalenus muscles[12] proved to be a fruitful method of diagnosis. These nodes were positive in 36 of 50 patients in whom the procedure was done. The biopsy was done usually on the right side; however, in 4 instances it was necessary to perform a biopsy on both sides to obtain a positive diagnosis. In 9 cases bilateral scalene node biopsies were negative, the diagnosis then being established by tissue removed from the lung or mediastinum at thoracotomy. The yield of positive biopsies at different sites is indicated in table 2.

Treatment: Patients presenting with mediastinal adenopathy alone or mediastinal adenopathy plus minimal parenchymal infiltration were given no specific treatment. During the last three years of the study, those patients who showed more than minimal pulmonary parenchymal infiltration were treated with adrenal corticoids. The usual program consisted of prednisone, 60 mg. per day, for several days and then a maintenance dosage of 30 mg. per day for a period of six to eight weeks. The drug was then gradually withdrawn. If the tuberculin test was positive, isoniazid and para-aminosalicylic acid were given prophylactically during the corticoid therapy and continued for four weeks after corticoid therapy was discontinued.

Fifteen of the patients treated with adrenal corticoids were followed from three to twenty-one months (average, eleven months) after initiating treatment. There was complete clearing of the parenchymal infiltration and disappearance of the enlarged mediastinal lymph nodes, as judged by roentgenogram, in 3 cases. The improvement was moderate in 6, and the abnormal shadows remained essentially unchanged in 6 additional cases. Although the clearing was occasionally rapid and dramatic, the part played by corticoids is not known. In 7 of the untreated patients who were followed for a comparable time, the abnormalities cleared completely in 2 and in 5 remained the same. A majority of the patients were returned to duty. Nine were discharged to limited duty and were re-examined after six months. In none of these was there recurrence or evidence of progression of the disease at the time of re-examination. Six patients were separated from the Service because of physical disability attributable to sarcoidosis. Four were separated because of expiration of enlistment, but were otherwise fit for duty. There was one death attributed to central nervous system involvement.

* * *

The low incidence of involvement of the skin, eyes, parotid glands, and bones is in contrast to other reports in the literature.[2, 3] This may be due to the fact that more than half of the cases reported here were discovered by routine roentgenographic examination of the chest during an early stage of the disease. Annual photofluorographic examinations of the chest are made of Navy and Marine Corps personnel. It is possible that many of these cases will progress to more extensive involvement, although there was no evidence of this in the relatively brief period of follow-up study. It is suggested that sarcoidosis

[12] DANIELS, A. C.: Dis. Chest, 1949, *16*, 360.

may be more prevalent than is generally recognized. Certainly, some of the cases reported here would have gone unrecognized if routine roentgenograms had not been taken. The roentgenograms in 5 cases cleared completely, and no detectable stigmata of the disease remained.

With the exception of the involvement of the central nervous system, the main cause of morbidity in sarcoidosis results from involvement of the parenchyma of the lungs. If this could be reversed by any modality, it would be desirable. Although the present study lends little concrete support to the value of adrenal corticoids, the clinical impression was gained that there was more rapid clearing of the parenchymal disease in the lungs of those patients so treated. It seems reasonable that the chance of fibrotic complications may be reduced by treatment of pulmonary infiltrates early in the course of the disease.

The relationship of tuberculous infection to sarcoidosis is by no means clear. Pinner reported that caseating tuberculosis was found in 14 of 44 cases of sarcoidosis at autopsy.[12] One of the patients not included in this series showed the typical granulomas of sarcoidosis as well as caseation necrosis containing tubercle bacilli in a single lymph node. Adjacent lymph nodes showed only sarcoid lesions and no tubercle bacilli by smear or culture. This patient had a positive skin test, using intermediate-strength PPD tuberculin. Long-term antituberculous therapy was given and the patient made an uneventful recovery. It is believed that active tuberculous disease cannot be excluded in those cases of sarcoidosis presenting positive tuberculin tests. If corticoids are to be used, then covering antituberculous drugs should be administered during and immediately following the course of adrenal corticoid therapy.

In summary: The cases of 59 patients with sarcoidosis admitted to a naval hospital between 1947 and 1958 have been reviewed. Sixty-three per cent of the cases were discovered by routine roentgenograms of the chest.

Mediastinal adenopathy was present in 88 per cent and parenchymal infiltration of the lungs in 54 per cent. The importance of a scalene lymph node biopsy in the diagnosis is shown by the fact that 72 per cent of 50 cases so examined were positive for sarcoidosis.

During the latter part of the study period, it was the policy to treat those patients who had more than minimal infiltration of the lungs with adrenal corticoids. The period of treatment was six to eight weeks. Antituberculous chemotherapy was administered prophylactically when the tuberculin skin test was positive. It was the clinical impression that those patients treated with adrenal corticoids showed more rapid resolution of the parenchymal lung disease than those untreated.

Only 6 of the 59 patients required separation from the Service because of physical disability resulting from sarcoidosis. Except for one fatal case, due to sarcoid involvement of the brain, there was no case which showed progression of the disease during the period of observation.

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June 12, 1959

[12] Pinner, M.: Pulmonary Tuberculosis in the Adult, ed. 2, Charles C Thomas, Springfield, Illinois, 1946.

LEGAL CUSTODY OF RECALCITRANT TUBERCULOUS PATIENTS

Report of a Hospital Program

In 1955, the General Assembly of the State of Ohio enacted legislation authorizing legal hospital custody for recalcitrant tuberculous patients. The procedure requires that a complaint be filed by the health commissioner in the patient's home district. The probate court of his county hears the case after adequate legal notice to the patient. If the complaint is upheld, the patient is transported to the detention unit of the Ohio Tuberculosis Hospital by a sheriff's deputy. He remains under court order until this order is lifted by the probate judge who issued it. The hospital administration has no responsibility for determining whether a patient should be admitted or for determining the proof of "communicability" of tuberculosis on which the court makes its decision.

The hospital does have the responsibility, however, of recommending discharge when the period of communicability has passed. This becomes a matter of medical judgment, since the decision is based on more than the presence or absence of tubercle bacilli in the sputum. The concentrated sputum smear must be negative for acid-fast bacilli but, also, previously visible cavities seen on roentgenographic examination must be closed and a sufficient period of time must have elapsed after the above changes to indicate that the patient is safe for contact with others.

The average occupancy of the unit is about 10 patients and has been as high as 17. From the beginning, it has been the policy of the hospital to provide as much in the way of necessary and desirable treatment and patient activities as is furnished to other patients, providing these are consistent with the operation of the detention unit. The staff is indoctrinated with the idea that it is not its purpose to punish anyone, no matter what his previous record may be. The unit is quiet, no force is required, and patient cooperation is usually obtained.

The usual patient arrives full of anger and resentment, expecting the worst. After a week or so, the nature of the hospital adjustment becomes more apparent. Individual patients, although reacting differently, generally show an adaptation that could be characterized as one of sullen cooperation. Only in rare instances are there gross difficulties so that limits have to be set, isolation for security used, or some form of discipline considered. Although these techniques are available, they are generally thoroughly discussed and applied only after a staff consensus has been obtained.

Newly admitted patients pass through a period of orientation to, and education about, their tuberculosis. Many of them have previously acquired incorrect information. If their behavior is reasonably good and if they show an understanding of their problems, they will be considered for transfer to an open ward after a period of several weeks. Decisions are made at weekly meetings of the staff committee appointed for this purpose. This committee represents several specialties, including occupational therapy, social service, medicine, and nursing. The progress and status of each patient on the unit is considered individually, and recommendations are made for future management.

Each area which operates a detention unit for recalcitrant patients will have its own problems which cannot be defined in advance. Some of them can be anticipated and prepared for; others will be learned and resolved only by experience.

This hospital's experience suggests that a maximal number of 15 to 17 patients of this type is desirable for efficient medical, psychiatric, and social management. One could easily overload a facility, invite difficulties, and ruin the morale of patients and personnel. It is believed that it would be unwise to proceed with the operation of a detention unit without having facilities and personnel to carry out the program in a satisfactory way.

Not all patients accept the hospital regimen. A number refuse to observe the rest and exercise schedule of the detention unit. Since the policy is not to force treatment, no discipline is attached to such behavior so long as other patients are not disturbed. Such lack of cooperation, however, would ordinarily tend to prevent transfer of the patient to a regular ward.

For the patient who is beyond persuasion or who is abusive or threatening, a brief stay of twenty-four or forty-eight hours alone in a security room with simple equipment has been effective and has increased the degree of cooperation. Seven patients have been subjected to this discipline. It has been learned that patients who are borderline psychotics and those who are full of resentment and tension will respond at certain times *only* to this sort of management. In 2 or 3 patients, there was strong evidence that they were

TABLE 1

OHIO TUBERCULOSIS HOSPITAL COURT ORDER PATIENTS

Admitted 11/4/57 to 5/4/59			50 patients
Men	41	White	44
Women	9	Negro	6
Age range			19 to 78 years
Average age			45.5 years
Previous hospital admissions			0–7
Average			2.12
Previous medically unsanctioned discharges			0–7
Average			1.86
No previous tuberculosis hospital care			4

TABLE 2

EXTENT OF DISEASE AND DURATION OF TUBERCULOSIS PRIOR TO PRESENT HOSPITALIZATION

Tuberculosis classification on admission	50 patients
Minimal	4
Moderately advanced	18
Far advanced	27
Tuberculo-silicosis	1
Duration of tuberculosis: Range	from 4 to 264 months
Average	49.8 months
Previous thoracic surgery: 5 cases—10%	

psychologically and socially maladjusted when transferred *to* an open ward. They acted in such a provocative manner as to require their return to the detention unit, which they welcomed!

One patient in the group discharged during this period refused to accept his antituberculosis drugs part of the time, and 2 patients refused surgical recommendations. An occasional patient remained uncooperative and hostile over a long period of time. Usually, such a patient is not transferred to a regular ward. A few individuals may display prolonged irritability because of being locked up and develop a better attitude only when transferred to an open ward. One court-order patient had left the hospital without authorization. She was on an open ward at the time and was apprehended and returned to the hospital two days later.

Patients who were admitted during the eighteen months under study are identified in table 1. Thirty-one of the 50 were men, and 44 of the 50 were of European descent. The average age was 45.5 years, with a range from 19 to 78. This table also records the number of previous hospital admissions and medically unsanctioned discharges.

The extent of disease in this group and the duration of tuberculosis prior to this hospitalization is shown in table 2. It is noted that 10 per cent had previously had some form of thoracic surgery in treatment of their tuberculosis.

Tables 3 and 4 deal with the 24 patients who were discharged during the period under study. The total duration of stay averaged 202 days. The time spent on the detention unit and on open divisions by this group is also displayed. It is noted that 12 (50 per cent) had no positive laboratory findings for tubercle bacilli during this admission. This was a surprising situation, since all patients were committed with the idea that they were community hazards and presumably infectious. A study of these 12 patients reveals, however, that 4 had definite open cavities, 3 more had filled cavities, 2 patients had had resections and were admitted for convalescent care after prior medically unsanctioned discharge, 2 patients showed nodular disease which regressed slowly just prior to and during hospital care, and the other patient showed noncavitary nodular disease which was unchanging and thought to be inactive. This patient had refused cooperation with his local health department so that his status was unknown prior to admission. He was promptly discharged when his inactive condition was understood. It appears that there was ample justifica-

TABLE 3

TYPES OF DISCHARGES, DURATIONS OF STAY, AND TYPES OF DETENTION

Discharged	24 patients
Regular	22
Psychiatric hospital	2
Duration of stay	41 to 413 days
Average	202.16
On detention unit	28 to 235 days
Average	95.25
On open division	0 to 302 days
Average	106.91

TABLE 4

LABORATORY AND DISCIPLINARY DATA ON THE 24 DISCHARGED PATIENTS

Tubercle bacilli in the sputum:		
"Positive" on admission	12	
Converted to "negative"		11
Transferred—still "positive"		1
"Negative" throughout	12	
Cooperation:		
Good—12		
Fair— 9		
Poor— 3		
Use of discipline:		
Yes— 7		
No— 17		

tion for admission for diagnosis and treatment of all of these patients.

For 4 patients, this was the first tuberculosis hospital care. These patients had refused cooperation with their local health departments. Enforced hospitalization seemed the only way to determine whether they had significant active disease. All of this group turned out to have active tuberculosis.

Classification of the recalcitrant patients into psychiatric categories was undertaken. This was done to separate the different syndromes and disorders and to study the relation of the patients and their recalcitrant status to their psychiatric classification.

Each patient was seen by a psychiatric resident. Subsequently and independently, he was seen by the consulting senior psychiatrist. Thus, there were two opinions, but few differences in classification occurred except in the subheadings or perhaps in those patients in the marginal group. Although an explanation was not sought routinely for the patients' previous refusal to accept hospitalization, this was always formulated by the senior psychiatrist.

A study of the 50 patients (table 5) shows that 2 were suffering from a psychosis with organic brain syndrome. One of these had such difficulties in orientation and created so many management problems that he had to be committed to the state mental institution for the tuberculous. In both of these patients, judgment was so impaired that they could not comprehend the need for hospitalization.

Three patients were diagnosed as chronic paranoid schizophrenics. One was committed to the state hospital because he continually fought with personnel, mostly with words, about regulations and taking his drugs. Another refused drugs saying, "It is against God's will"; but, because he was cooperative in other respects, he was not committed to a psychiatric hospital. The third schizophrenic patient who had an alcoholic history went through many verbal assaults but, because no physical assaults were expected or occurred, he finished his treatment program. These 3 patients could not accept hospitalization previously because they were all suspicious, suffered feelings of isolation from their communities, and led a schizophrenic way of life which, they believed, was threatened by living in a tuberculosis hospital.

TABLE 5

PSYCHIATRIC CLASSIFICATION OF RECALCITRANT PATIENTS

Psychosis with organic brain syndrome	2
Psychosis with schizophrenia, chronic paranoid	3
Alcoholism, chronic	16
Personality pattern disturbance	7
Marginal socio-psychiatric adjustment problem	12
Mental deficiency	1
Organic brain syndrome, mild, without psychosis	2
Miscellaneous	7
Total	50

By far the largest group was the alcoholic patients. As in all people classed as alcoholic, many problems, diagnoses, and social factors are known to exist. Sixteen of 50 patients were classed as chronic alcoholics. At least 7 of these patients were in acute alcoholic episodes at the time of admission, thereby giving an answer, in part at least, for not accepting hospitalization. All could be classed as having one of the personality disorders with alcoholism and, in addition, 2 or more were in borderline psychotic states or perhaps chronic undifferentiated schizophrenia. One 58-year-old male had evidence of a moderate organic brain syndrome, probably due to his thirty to forty years of alcoholism.

A group of 7 patients more difficult to explain were classed as personality disorders. It is known that patients so classified often *do* accept hospitalization, and it is puzzling how the personality structure of the patient, in itself, explains this recalcitrancy. It is considered that their family and social milieu produce a conflict, so that they have to deny or refuse to accept hospitalization lest their tenuous position in the family be uprooted.

Twelve patients seemed to elude strict psychiatric classification. They all had borderline or low intelligence quotients; they appeared to be inadequate, immature, or to have inferiority feelings; they had made marginal social adjustments so far as their jobs or family life was concerned. Their behavior was schizoid, antisocial, or dissocial. Since much of their behavior was on the *margin* of attaining adjustment, but seemed not quite to "make the grade," they were designated as "marginal personality." It seemed likely that they were the type of patients who would resist hospitalization because they believed they could not successfully adjust to the hospital.

One patient was in a class by himself because of his very low intelligence quotient, and was diagnosed as mentally deficient. He was also in trouble because of alcoholism, so that between his dependency needs and lack of understanding he could not comprehend the meaning of hospitalization.

Two patients were in the group of organic brain syndrome without psychosis. Their memories, judgments, and retention were slightly impaired. It seemed that, at the ages of seventy and seventy-eight, they believed they should not be required to change their ways and come to a hospital.

The remaining 7 patients comprised a miscellaneous nonclassifiable group, but were viewed as presenting problems peculiar to the individual. Two of these patients were "mother-attached," single, middle-aged men. Another was a patient with a thirty-five-year history of disabling rheumatoid arthritis and a nine-year history of chronic barbiturate dependency. One 43-year-old recently married female had her marriage interrupted by the diagnosis of tuberculosis and a concomitant mixed psychoneurotic syndrome. Another 63-year-old patient thought his rights to visit his family were limited by a tuberculosis hospital. He became recalcitrant and showed primarily a denial reaction of his tuberculosis. The sixth patient was a 58-year-old retired miner who believed that he could get treatment and hospitalization at this hospital only if he were discharged on a medically unsanctioned basis from his previous hospital. This he accomplished. A seventh patient was an active male, aged seventy-five, who by his aggressive, obstinate personality had resisted hospitalization for more than seven years. He even accumulated affidavits showing he had sputum negative for tubercle bacilli and maintained his denial of tuberculosis by projection, accusing others for his hospitalization.

* * *

It is reasonable to infer that each patient has his own rationalization for refusal to accept hospitalization, and in a few cases this did seem plausible. In the light of their psychiatric condition, socio-psychiatric adjustment, and personality structure, most of the patients provide a deeper understanding of their recalcitrancy. With these considerations in mind, the patients were studied according to their psychiatric classification, and some inferences were drawn. These inferences led to generalizations which have dynamic and theoretic value. If they are applied too rigidly in any one case or overvalued as the most important factor in explaining recalcitrancy, they might be misleading. There are many factors in each case which may be of equal significance. Since this report is concerned with a sample population small in number and early in the operation, it was thought to be wise to study the patients in more general terms in order to test or further evaluate our understanding in future studies.

Of 50 patients, 21 had been arrested one or more times for drinking. Twelve had served one or more prison sentences for other causes, mostly minor; only 2 of these had not been arrested for alcoholism also. These experiences, along with their personal problems, would tend to make those with prison records avoid further seclusion in a hospital. Up to now no patients in this unit have been narcotic addicts. Certainly, these individuals would complicate management problems a good deal.

Many alcoholics become almost model patients with hospitalization because of the great dependency needs which the hospital fulfills. At times this is not the case when the system of paranoid projection or authority problems becomes more prominent during hospitalization. Even though alcoholism is a common reason for this group of patients to resist voluntary hospitalization, it does not explain why some alcoholics accept hospitalization at times and at other times become recalcitrant. When alcoholism is a factor, repeated hospitalizations can be expected, not only for tuberculosis, but also for mental illness and any other chronic or recurrent ailment.

Some interesting assumptions are possible from this study. Alcoholism is the most frequent factor in recalcitrancy, but not nearly so frequent as in the study at Firland Sanatorium. That sanatorium found alcoholism the main factor in 69 of 82 reported patients, whereas we found it in 16 of 50 cases. They classified these as "Skid Row" patients, but the nearest we could come to this classification was the "marginal personality." It is possible that the alcoholics' personality disorders and Skid Row patients at Firland represent the migrant type of population of the state of Washington and, in particular, Seattle. Whereas it is likely that in Ohio, with its inland cities and rural population, the problem is not as great and manifests itself in more subtle ways. Thus, we looked for Skid Row or its equivalent in our patients and could not find it. Instead, we found and postulated a marginal socio-psychiatric personality problem type. It may be assumed that hospitalization is a socializing force that is resisted, resented, and rejected by the socially maladjusted person from the marginal and the Skid Row groups.

Tuberculosis brings this factor of resisting the socializing force of hospitalization into the open for these persons. Conclusions by other writers suggesting dependency conflicts, psychologic incompatibilities, and family difficulties as the determining factors in recalcitrancy are not an adequate explanation. Individual appraisal is

important because no one factor or combination of factors can explain all recalcitrant patients.

In summary: The background and some of the policies and attitudes applied in the operation of a unit for the compulsory hospitalization of recalcitrant tuberculous patients have been presented.

Requirements for the effective operation of a unit for recalcitrant tuberculous patients include adequate physical facilities, a sufficiently well-indoctrinated staff, carefully delineated written policies, a therapeutic nonpunitive attitude, a small detention unit with provision for open division treatment, frequent individual consideration of patient problems and progress, and routine psychiatric consultation. Adequate care for this group is expensive, but it can be accomplished without major upset of operation or morale of the hospital.

The study of these 50 patients revealed a variety of psychiatric problems with a significant number of chronic alcoholics and marginal sociopsychiatric personalities. Treatment results indicated that a large percentage were suitable for effective tuberculosis therapy and had a fairly good long-term prognosis.

It is believed that the public health requires this legal custody and that in most instances it benefits the patient. In the past eighteen months, forcible hospital care of recalcitrant patients has shown that it is important in the control program of tuberculosis. Nevertheless, further evaluation and research are in order.

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June 15, 1959

Books

THE CARE OF THE GERIATRIC PATIENT. Edited by E. V. Cowdry. Pp. 438, C. V. Mosby Company, St. Louis, 1958. Price, $8.00.

This interesting manual, written by twenty-two contributors, makes it abundantly clear that the care of the elderly is an increasing responsibility of all doctors and can also be interesting to the busy practitioner. The contributors are not unanimous in settling the question whether geriatrics is a specialty or not, but all agree that it is a mixture of clinical practice plus attention to nonmedical aspects of patient care.

The chapter on psychologic aspects in the management of the geriatric patient is a very clear-cut exposition of some of the problems most aggravating to the physician in tending the elderly.

The concept that "disuse" atrophy of the brain may explain some of the mental problems of the aged gives a practical clue for prevention. It is also heartening to note that pathologic changes in the brain do not necessarily bear a one-to-one relationship to levels of performance.

The chapter on medical aspects of geriatric care is particularly challenging. It states that the chronic diseases of the elderly are the transient functional diseases of youth, and that cancer at 60 is the tissue change at 40. If this is true, geriatrics is the specialty of treating preventable diseases.

The chapter on drugs and the treatment of the geriatric patient urges that more time be spent in safeguarding persons by listing the things not to do. Time, rather than prompt action, is the thing required of the doctor in geriatrics.

In the chapter on hospitalization of the geriatric patient, a strong argument is made for the voluntary general hospital to expand its facilities to this end. This is much preferable to the hospital that limits itself to the care of the elderly, which is "too often among the saddest spots in creation." Coupled with the general hospital should be a good home-care program and rehabilitation. It is stated: "to employ a full team in every patient is to be unreasonably inflexible and to misuse ones' resources." This is particularly true in the rehabilitation of the elderly. Logic is the best basis of treatment and spontaneous recovery in hemiplegia "must be given the major share of credit."

Continually throughout the book the importance of educating doctors and paramedical personnel to the role of rehabilitation is mentioned. In Chapter 17, a possible scheme for teaching geriatrics in medical schools is outlined, but I have grave doubts about the effectiveness of a coordinator working under a committee made up of department heads.

The chapter listing available organizations and services for older people should be carefully read by all who practice medicine. Cooperation with these agencies is essential, and yet they are too infrequently used.

This is a book that attacks the problem very realistically, and its use would make the treatment of elderly patients more interesting to any practitioner. It is extremely readable and is well indexed.

HAROLD N. WILLARD
Waterville, Maine

LES HÉMOPTYSIES TRACHÉO-BRONCHIQUES. By A. Meyer and J. Chrétien. Pp. 158, Masson et Cie, Paris, 1958.

This book represents the sum of experience acquired these past years by the authors in a very active chest service of one of the Paris hospitals (Boucicaut), regarding one key-symptom: hemoptysis. The statistics include 717 cases in which hemoptysis was the initial symptom. In an important number of the cases, by bronchoscopic examination it was possible to demonstrate the bronchial origin of the hemorrhage. This is the point that the authors want to emphasize. The different possible causes of the alarming symptomatology are thoroughly and authoritatively discussed, with the support of a conscientious bibliography. But if one goes to the trouble of writing a book and reviewing 40,000 patient charts, why not make the book more appealing by completing the text with good endoscopic pictures related to the different pathologic entities and, perhaps, some characteristic chest roentgenograms? The only photographs are rather poor reproductions of histologic slides.

PIERRE E. FERRIER
Seattle, Washington

LES MALFORMATIONS CONGENITALES DU POUMON. By A. Breton and O. Dubois. Pp. 140, Doin Editions, Paris, 1957.

This is an intelligent and easily readable monograph on a difficult subject which has acquired

some importance since the modern development of pulmonary surgery and bronchoscopy. It deals essentially with the embryologic, anatomic, and clinical aspects of the problem. Therapy is only briefly outlined. The roentgenograms are, in general, satisfactorily reproduced. No overemphasis is placed on the classification of these various anomalies and this is well, since no classification can be proved to be better than another in such a matter. This is especially true for the hypoplastic lung, or the infantile bullous pneumopathy, for instance.

The whole book, chapter by chapter, is built on a rather thorough bibliography, which shows that the authors are familiar not only with the French literature, but also with the Anglo-Saxon, German, Italian, and Spanish. (This contrasts with a recently published French monograph on asthma in childhood, which contains only 17 references, all French.) We have been surprised, however, to see the authors indulge in such language offenses as "un mécanisme de check-valve," or "blebs sous-pleurales."

The lack of histologic illustration is to be deplored, even though histology does not bring up more than uncertainties as far as the origin of these anomalies is concerned.

PIERRE E. FERRIER
Seattle, Washington

LA CORTICOTHERAPIE SURRENALE DANS LE TRAITEMENT DE LA TUBERCULOSE. By Ch. Gernez-Rieux, H. Warembourg, and M. Pauchant. Pp. 176, Masson et Cie, Paris, 1958.

This monograph is the result of four years of clinical experience in the use of corticosteroids in more than 500 tuberculous patients. After an historical introduction and critical analysis of the reasons for the initial opposition to the use of corticosteroids in tuberculosis (overdosage and omission of adequate coverage with antimicrobial drugs), the authors discuss in great detail the results which they and others have obtained in the application of this therapy in the various forms of tuberculosis, such as the primary complex, acute and chronic pulmonary tuberculosis, bronchial tuberculosis, involvement of the serous membranes, and tuberculous meningitis. The roles of the corticosteroids in the treatment of patients with allergic reactions to antimicrobial drugs in cases which require surgical intervention and in the treatment of other diseases associated with tuberculosis, such as asthma, rheumatism, and cirrhosis of the liver, are also discussed. Other chapters deal with the effect of corticosteroids on tuberculin allergy and other biologic tests of tuberculous infections, and with the untoward incidents and accidents which may occur under corticosteroid therapy. The literature concerning corticotropin and cortisone in experimental tuberculosis is also analyzed, and the authors present their own investigations on the mode of action of these hormones in clinical tuberculosis.

The benefits from corticosteroid therapy are often remarkable. It is emphasized that only patients with bacilli susceptible to the antimicrobial drugs, particularly isoniazid, should be selected for treatment with steroids and that the steroids should be used only in combination with prolonged antimicrobial therapy.

The book is written in a pleasant style. Case reports are accompanied by roentgenograms. The literature is exhaustively reviewed and often advantageously summarized in the form of tables. The bibliography contains more than 500 references. The book is highly recommended.

WERNER B. SCHAEFER
Denver, Colorado

BOOKS RECEIVED

CIBA FOUNDATION STUDY GROUP NO. 1: PAIN AND ITCH: NERVOUS MECHANISMS. Edited by G. E. W. Wolstenholme and Maeve O'Connor. Pp. 120, Little, Brown and Company, Boston, 1959. Price, $2.50.

CIBA FOUNDATION STUDY GROUP NO. 2: STERIC COURSE OF MICROBIOLOGICAL REACTIONS. Edited by G. E. W. Wolstenholme and Cecilia M. O'Connor. Pp. 115, Little, Brown and Company, Boston, 1959. Price, $2.50.

DIAGNOSTIC ET TRAITEMENT DE LA TUBERCULOSE PULMONAIRE. By J. Centner. Pp. 235, Éditions Arscia S. A., Bruxelles, 1959.

CLINICAL EVALUATION OF NEW DRUGS. Edited by S. O. Waife and Alvin P. Shapiro. Pp. 223, Hoeber-Harper Inc., New York, 1959. Price, $7.50.

BRONCHIAL ASTHMA. A Symposium. Pp. 96, Chest and Heart Association, London, 1959. Price, 12s. 6d.

ROBERT KOCH. By Wilhelm Kolle. Pp. 67, Georg Thieme Verlag, Stuttgart, Germany (in U. S. and Canada, Intercontinental Medical Book Corporation, New York), 1959. Price, $4.30.

American Trudeau Society

Medical Section of the National Tuberculosis Association

NOTICES

The following dates and locations of the next five annual meetings of the National Tuberculosis Association and the American Trudeau Society have been selected and approved:

1960—Los Angeles, California—May 15–20
1961—Cincinnati, Ohio—May 21–26
1962—Miami Beach, Florida—May 20–25
1963—Denver, Colorado—May 12–17
1964—New York, New York—May 24–29

1960 ANNUAL MEETING OF THE NATIONAL TUBERCULOSIS ASSOCIATION

Plans are now being made for three days of scientific sessions at the annual meeting of the National Tuberculosis Association next May in Los Angeles, California. The scientific sessions will be sponsored by the Association's medical section, the American Trudeau Society, and will be held on May 16, 17, and 18, 1960.

The American Trudeau Society meetings will be held in conjunction with the annual meeting of the National Tuberculosis Association, which will open May 16 and extend through May 19. Public health sessions will be held concurrently with the medical sessions. Scientific exhibits will be on display throughout the meeting. The Statler-Hilton and the Biltmore will be headquarters hotels.

Papers will be selected from abstracts submitted to the program committee not later than January 7, 1960. Abstracts should be sent to David W. Cugell, M.D., chairman of the Medical Sessions Committee, American Trudeau Society, 1790 Broadway, New York 19, New York.

In addition to sessions at which papers will be presented, there will be five panel discussions and seven luncheon seminars. The panels will be on the following topics: Childhood Tuberculosis, Tissue Culture Techniques and Mycobacterial Research, Steroids in Nontuberculous Pulmonary Diseases, Adenoviruses in Respiratory Diseases, and the Treatment of the Chemotherapy Failure. The seminars will be on the following subjects: Practical Pulmonary Function Testing, Treatment of Bone and Joint Tuberculosis, Compulsory Isolation of the Recalcitrant Patient, Air Pollution and Pulmonary Diseases, Indications for Lung Collapse, Surgery of Lung Cancer, and Treatment of Empyema.

POSTGRADUATE COURSE ON DISEASES OF THE CHEST

The Council on Postgraduate Medical Education of the American College of Chest Physicians will present the 13th Annual Postgraduate Course on Diseases of the Chest at the Sheraton Hotel, Philadelphia, March 14–18, 1959.

The most recent advances in the diagnosis and treatment of heart and lung diseases, medical and surgical aspects, will be presented. Tuition for this five-day course will be $100, including round table luncheon discussions.

Further information may be obtained by writing to the Executive Director, American College of Chest Physicians, 112 East Chestnut Street, Chicago 11, Illinois.

THE AMERICAN REVIEW
OF RESPIRATORY DISEASES

Clinical and Laboratory Studies of Tuberculosis and Respiratory Diseases

ABSTRACTS

Edited by EDWIN D. KILBOURNE

VOLUME 81 February 1960 ABST. No. 2

The Editor recognizes the inevitable incompleteness and arbitrary selectivity of any abstracting service, including this one. It is his belief that the usefulness of the present Abstracts section will be enhanced if the reader is informed of the general purpose of the abstracts and the criteria employed in their selection.

It is the purpose of the Abstracts section to acquaint the diversified and international readership of the REVIEW with the gist of original and scientific articles which are related to the general subjects of tuberculosis or respiratory diseases implicit in the title of the journal. In this connection, 40 abstracters systematically review 207 journals which represent 31 countries and which are published in 18 languages. In most instances, abstracters cover journals in fields of their special competence. It is evident that the diverse interests of the readers of the REVIEW dictate a balanced selection of clinical, laboratory, and epidemiologic studies. Such selection will often preclude the publication of articles equal in scientific merit to many presented here.

In the publication of abstracts, cognizance is taken of the extent to which the journals yielding them are circulated. Articles from journals of wide circulation may be mentioned by title only. This space-saving device is also employed for articles which defy condensation without the omission of essential data or discussion. The attention of readers is thus directed to the existence of papers which are no less important because they are not summarized.

(Requests for further information on abstracted articles should be directed to the author of the article at the address indicated by an asterisk (*) in each abstract title.)

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NONTUBERCULOUS THORACO-PULMONARY DISEASE

Nontuberculous Mediastinal Adenopathies with Erythema Nodosum (Löfgren's Syndrome). Four New Case Reports (in French). A. MEYER,* M. RAUGEL, J. L. JULLIEN, and A. NADOLNY. *Rev. tuberc., Paris*, April, 1959, 23: 357–372. (*Paris, France)

The association of nontuberculous mediastinal adenopathy and erythema nodosum was first described in the Scandinavian literature by Löfgren in 1946 and 1952. Four additional cases of this syndrome are described. All showed onset of illness with fever and erythema nodosum; this was associated with pain and swelling of multiple joints. Tonsillitis was observed in 2 cases. The tuberculin reaction was negative in 2 cases, in 1 of which it was known to have been positive before the illness. All patients showed massive mediastinal adenopathy. Three of the 4 cases subsequently developed parenchymal nodulation. The response to steroid therapy was good in all patients. The diagnosis of sarcoidosis would seem to be the most probable one; however, the onset of illness was typically that of an infectious disease; this, as well as the joint manifestations and tonsillitis are not usually seen in sarcoidosis. The antistreptolysin titer was 100, 200, and 500 in 3 patients. Blood cultures for streptococci were negative.

V. LEITES

Dissecting Aneurysm of the Thoracic Aorta. F. KNUTSSON.* *Acta radiol.*, April, 1959, 51: 273–277. (*University Hospital, Upsala, Sweden)

The underlying causes and mechanisms of the development of a dissecting aneurysm are given. The so-called classical form and the arteriosclerotic form are discussed. The onset of symptoms is described as being dramatic with severe constrictive pain in the back, thorax, or abdomen. The patient is usually distressed and anxious. The clinical picture varies with the localization of the lesion. The onset may develop gradually without marked symptoms. Because of the increasing tendency toward operative intervention, the diagnosis is becoming increasingly important. Roentgen examination and aortography are discussed as an aid in the diagnosis. Two case reports are given in some detail.

E. SHABART

Red-cell Aplasia and Benign Thymoma. E. H. O. PARRY,* G. S. KILPATRICK, and R. M. HARDISTY. *Brit. M. J.*, May 2, 1959, No. 5130: 1154–1156. (*Royal Infirmary, Cardiff, Wales)

A case of benign thymoma and red-cell aplasia is described. Thymectomy did not produce a hematologic remission, but normal erythropoiesis occurred later during treatment with prednisone. The patient's plasma did not inhibit erythropoiesis when injected into normal rats (Authors' summary).

E. A. RILEY

Pathogenesis of Arrhythmias Associated with Metastatic Tumors of the Heart. T. N. JAMES* and G. M. CARRERA. *New England J. Med.*, April 23, 1959, 260: 869–871. (*Tulane U. School of Med., New Orleans, La.)

In a case of bronchogenic carcinoma with solitary metastasis to the heart, 3 lesions were found: neoplastic invasion of a sympathetic nerve in the atrium, an atrial coronary artery with subsequent thrombotic occlusion and atrial infarction, and noninfarcted atrial myocardium. The possible relation is discussed of these 3 lesions to the atrial arrhythmias that were a problem in the management of the patient. It seems reasonable that lesions analogous to the 3 discussed may also be the basis for "spontaneous" atrial arrhythmias in older patients, except that they are due to focal atherosclerosis instead of neoplasm.

M. J. SMALL

Gamma Globulin in the Management of Asthma Associated with Infection. S. FRIEDLAENDER,* A. S. FRIEDLAENDER and L. WEINER. *Am. J. M. Sc.*, July, 1959, 238: 18–23. (*Wayne State U. College of Med., Detroit, Mich.)

Pooled γ-globulin in doses of 0.3 to 0.5 ml. per kg. of body weight was administered prophylactically every 3 weeks for periods of 8 to 12 months to 15 children, aged 5 to 11, with so-called "infectious asthma." Some reduction in the frequency of upper respiratory infections and subsequent episodes of asthma was noted in 8 patients during the periods of treatment. Examination of pre-treatment sera by paper electrophoresis was normal in all but 2 cases. Pre-treatment immunochemical studies of these sera revealed low levels of γ-globulin in 4 cases, high levels in 2 others. Post-treatment sera showed elevation in β-globulin fractions in almost all instances. Levels of γ-globulin were increased in 6 and diminished in 3 cases following treatment. There is a suggestion from this study that the results of prophylactic administration of pooled γ-globulin in such asthmatic children may be more favorable when a diminution in serum γ-globulin level exists before treatment.

W. J. STEININGER

A Psychosomatic Aspect of Asthma. J. A. CROCKET.* *Brit. J. Dis. Chest*, April, 1959, 53: 128–141. (*Glasgow Northern Hosp., Scotland)

The relationship of etiologic factors in bronchial asthma is discussed with particular reference to

the unity of the psychologic and somatic aspects. The relative importance, in a series of patients, of the psychologic factor, of respiratory infection, and of allergy has been shown to vary at different ages and between the sexes. The causes of the higher figures for the allergic factor in men, and also the curves taken both by this factor and by the psychologic factor in men, are less obvious. Probably the patterns are the outcome of several trends such as increased exposure to allergens at work, the higher incidence of bronchitis in men and, in later life, the appearance of emphysema with increasing frequency and severity.

M. J. SMALL

Impaired Pulmonary Function in Acute Beryllium Poisoning (in Japanese). T. MOMOSE,* S. KOIKE, A. SAKAMOTO, T. SHIRAISHI, E. YOKOYAMA, M. MURAO, and T. NEGISHI. *Nihon Rinsho*, July, 1959, 17: 1229–1237. (*Tokyo U. School of Med., Japan)

Acute berylliosis with respiratory impairment was seen in 24 cases for the first time in Japan. Two weeks to 3 months after exposure to beryllium, the patients developed signs and symptoms of upper respiratory infection, bronchitis, or pneumonitis which persisted for 1 to 9 weeks. Roentgenographic abnormalities were also reported in some cases. Pulmonary function tests revealed that the essential feature of the pathologic malfunction in these patients was impaired pulmonary diffusion. This conclusion was drawn from the following results: reduced vital capacity in many cases; normal residual volume; increased minute ventilation, in some cases with dyspnea; normal maximal breathing capacity and timed vital capacity except in severe cases; greater decreased arterial oxygen saturation during exercise than at rest in some cases; considerably increased alveolar-arterial oxygen tension difference in a severe case; arterial blood saturation reached its highest value when pure oxygen was breathed; and, normal arterial carbon dioxide tension, pH, and buffer base. These impairments improved gradually and left no clinical abnormalities even in the most severe case when studied 4 months later.

I. TATENO

Bronchiolitis. S. ENGEL. *Brit. J. Dis. Chest*, April, 1959, 53: 125–127.

Bronchiolitis may occur as general involvement of the bronchioli and thus cause a serious condition. This kind of bronchiolitis is rare and affects mainly very young children. Mural bronchiolitis may be primarily widespread or may be part of inflammation throughout the bronchial tree, especially in cases of fibrocystic disease of the pancreas. Localized and sometimes obliterative bronchiolitis is, as a rule, less serious.

The roentgenogram shows widespread "mottling" that resembles miliary tuberculosis or other diffuse conditions. The mottling, however, consists of fairly soft opacities, different in density from those of miliary tuberculosis. It is a diffuse mottling which may occupy the best part of both lung fields, especially the lower parts, and is so characteristic that it is of diagnostic value. Bronchiolitis may turn into a chronic condition. What causes acute bronchiolitis to turn into "bronchiolitis obliterans" is not known, but the author has often seen it in generally indurated lung tissue.

M. J. SMALL

The Coexistence of Primary Lung Cancer and and Other Primary Malignant Neoplasms. C. G. MOERTEL,* H. A. ANDERSEN, and A. H. BAGGENSTOSS. *Dis. Chest*, April, 1959, 35: 343–347. (*U. of Minnesota, Rochester)

A solitary pulmonary lesion in a patient having a proved primary malignant neoplasm in another site presents a crucial problem in differential diagnosis. The fact that a presumably metastatic lesion may in fact be a second and potentially curable primary malignant lesion must always be kept in mind. The fact that the lung is a common site for almost all types of metastatic malignant lesions compounds the difficulty of establishing a pulmonary lesion as a true second primary cancer.

Over a 10-year-period (1944–1953) a total of 1,588 patients of the Mayo Clinic had the diagnosis of primary cancer of the lung, bronchus, or pleura confirmed by tissue examination; of this group, 65 patients (4.1 per cent) were found to have one or more other primary malignant neoplasms. Of these patients, 59 were male and 6 were female. The distribution of the specific types of second primary cancers does not seem significantly different from that to be expected in random selection of patients of comparable age and sex with single malignant neoplasms.

E. A. ROUFF

Primary Alveolar Carcinomatosis of the Lung (So-called Pulmonary Adenomatosis or Alveolar-Cell Tumor). E. ROELSEN,* T. LUND, T. SØNDERGAARD, B. MØLLER, and A. MYSCHETZKY. *Acta med. scandinav.*, April 15, 1959, 163: 367–384. (*Central Hosp., Silkeborg, Denmark)

Twelve cases are reported, 8 of the multiple or diffuse form of primary alveolar carcinomatosis of the lung, and 4 of the solitary form (carcinoma). The clinical pictures of these two varieties were widely different; dyspnea and cyanosis were predominant in the diffuse variety but scarcely present in the solitary form whereas, with a few excep-

tions, a productive cough was common to both. Clinically, the most important criterion was the roentgenographic appearance, which in the diffuse form was highly variegated and difficult to interpret. Conditions which require differentiation are tuberculosis, sarcoidosis, bronchopneumonia, pneumoconiosis, bronchiectasis, blastomycosis, and coccidioidomycosis. The appearance in the solitary form was less complex, a solitary infiltration or round shadow, but also noncharacteristic.

Only 1 of the patients with the diffuse form raised abundant amounts of sputum. Cytologic studies of the sputum were performed in 8 cases, and revealed tumor cells in 5. Bronchoscopy was performed in 3 of the 4 solitary cases, with positive findings in 2. Daniel's biopsy was performed in 2, but yielded a positive result in only 1 case (of the diffuse form). The erythrocyte sedimentation rate was elevated in only 7 of the 12 patients.

Of the 8 cases of the multiple or diffuse form, 6 were subjected to autopsy, while only partial autopsy was performed in 2. The 4 solitary carcinomas were confirmed by histologic examination of the specimens removed at operation. It is most important to emphasize that the macroscopic changes, on section, were reminiscent of grey hepatization of lobar pneumonia. Histologically, the alveolar structure was preserved, the alveolar walls being lined with cancer cells with a varying degree of differentiation. Invasive growth into the lymphatics was common, and metastatic lesions were found in half of the cases.

In addition to the alveolar changes, 7 of the patients also revealed changes in the epithelium of the bronchioles. It is likely that this special form of cancer may arise both from the alveolar epithelium and from the bronchioles, and that it may be of either multicentric or unicentric origin. Etiologically, it is of interest that 8 patients with multiple or diffuse lesions were either nonsmokers or practically so, while 3 of the 4 patients with solitary lesions were heavy smokers. The prognosis of the multiple or diffuse form was obviously very grave. All 8 patients died within 9 months after the onset of symptoms. The 4 patients with solitary lesions were treated surgically; 2 of these are alive, 5 years, and 16 months, respectively, after operation.

The frequency of primary alveolar carcinomatosis of the lung during the years 1949–1956 was calculated to be 3.5 per cent of that of all of the lung cancer cases notified from the Danish district concerned (population approximately 500,000). The disease was about equally frequent in both sexes.

E. DUNNER

Fatal Chylopericardium Caused by Hamartomatous Lymphangiomatosis. S. U. MILLER,* H. J. PRUETT, and A. LONG. *Am. J. Med.*, June, 1959, 26: 951–956. (*U. of California School of Med., San Francisco)

This report describes a fatal case of chylopericardium associated with an extensive hamartoma of the intrathoracic lymphatic channels. The diagnosis was finally established by determining the lipid composition of the hemorrhagic chylous effusion, and by the use of an orally administered lipotrophic dye. Left thoracotomy and ligation of many of the mediastinal lymphatics was unsuccessful in relieving tamponade, and cardiorespiratory insufficiency led to death. Post mortem findings revealed a congenital defect of the intrathoracic lymph vessels leading to extensive lymphangiomatosis and lymphangiectasia (Authors' summary).

T. H. NOEHREN

Spontaneous Hydatid Cyst Pneumothorax (in Italian). R. D'AMBROSIO,* F. BRUNI, and V. CAVALLI. *Lotta contro tuberc.*, November–December, 1958, 28: 1166–1188. (*Osp. S. Filippo Neri, Rome, Italy)

The incidence of spontaneous hydatid cyst pneumothorax in Italy is decreasing, primarily because early diagnosis of the specific disease is more frequent, with consequent rapid surgical treatment. Two cases of this serious syndrome in women 27 and 41 years old are presented and their description is accompanied by a general clinical and pathogenetic consideration of this complication of the disease.

I. ARCHETTI

Cardiopulmonary Insufficiency Associated with Myotonic Dystrophy. K. H. KILBURN,* J. T. EAGEN, and A. HEYMAN. *Am. J. Med.*, June, 1959, 26: 929–935. (*Duke U. Med. Center, Durham, N. C.)

A 31-year-old man with a long history of myotonic dystrophy was observed to have somnolence, cyanosis, Cheyne-Stokes breathing, and a cardiac arrhythmia. Cardiopulmonary function studies revealed a decrease in vital capacity and total lung volume and a reduction in maximal breathing capacity. In addition, hypercapnia, hypoxemia, and pulmonary hypertension were observed. It is suggested that dystrophy of the respiratory muscles and possibly of the myocardium produced in this patient the syndrome of alveolar hypoventilation (Authors' summary).

T. H. NOEHREN

Partial Eventration of the Left Diaphragm. An Unusual Case. E. LAUSTELA.* *Ann. chir. et gynaec. Fenniae*, Fasc. 2, 1959, 48: 218–225. (*U. of Helsinki, Finland)

An unusual case of partial eventration of the left hemidiaphragm is reported. In the center of the hemidiaphragm, there was a nearly circular area about 5 by 8 cm. in size which consisted merely of a thin membrane of connective tissue which ascended into the thoracic cavity almost to the level of the hilum, forming a sac which contained the spleen and a portion of the colon and peritoneum.

Because of the marked distress—dyspnea, constipation, and meteorism—the patient was operated on. After reposition of the abdominal organs, the membrane was resected and the muscular margins of the diaphragm were sutured. The symptoms disappeared after the operation.

J. HAAPANEN

A Comparison of Sweat Chlorides and Intestinal Fat Absorption in Chronic Obstructive Pulmonary Emphysema and Fibrocystic Disease of the Pancreas. J. A. WOOD,* A. P. FISHMAN, K. REEMTSMA, H. G. BARKER, and P. A. DI SANT' AGNESE. *New England J. Med.*, May 7, 1959, 260: 951–957. (*Presbyterian Hosp., N. Y., N. Y.)

The concentration of chloride in sweat and the intestinal absorption of neutral fat and fatty acid were compared in adults with chronic pulmonary emphysema and in children with fibrocystic disease of the pancreas. Five of the 24 patients with emphysema had abnormally high concentrations of chloride in sweat, and 4 of these also had impaired absorption of neutral fat.

The sweat chloride in the emphysematous subjects reverted to normal during severe salt restriction. This pattern of response of the emphysematous subjects differed from that of children with cystic fibrosis of the pancreas, but was similar to that of the parents of these children. The observations suggest that some cases of chronic obstructive pulmonary emphysema in adults may represent *formes frustes* of fibrocystic disease of the pancreas (Authors' summary).

M. J. SMALL

Unusual Cases of Pulmonary Arteriovenous Fistulas, with a Note on Thyroid Carcinoma as a Cause. J. A. PIERCE,* W. P. REAGAN, and R. W. KIMBALL. *New England J. Med.*, April 30, 1959, 260: 901–907. (*U. of Arkansas School of Med., Fayetteville)

Multiple small pulmonary arteriovenous fistulas may be either congenital or acquired. Previously described patients with acquired fistulas have had either cirrhosis of the liver or pulmonary schistosomiasis, or both. This report adds metastatic thyroid carcinoma as a cause of multiple small pulmonary arteriovenous fistulas. Certain physiologic procedures in the assessment of venous fistulas are discussed, and 2 unusual cases presented.

The first patient had in the lungs numerous metastatic nodules of thyroid carcinoma that functioned as pulmonary arteriovenous fistulas. He had marked cyanosis. No cardiac abnormality was observed on physical examination, and none was demonstrated at cardiac catheterization or by angiocardiography. Thus, the heart was excluded as the site for this shunt flow. Although the possibility remains that multiple pulmonary arteriovenous fistulas existed on a congenital basis, this seems unlikely. In the first place, no angiomas were found on careful inspection of the skin and mucous membranes. Secondly, the observed chest lesions actively absorbed radioactive iodine. Finally, the pulmonary symptoms appeared only a few months before the diagnosis of thyroid carcinoma was made. Because the arterial hemoglobin oxygen saturation increased after therapy with radioactive iodine, it is reasonable to conclude that the shunt flow occurred through vessels supplying the tumor metastases in the lungs. Whenever possible, pulmonary function studies should be performed in patients with pulmonary metastases from thyroid carcinoma before any treatment is begun.

In the second case, although abnormal vascular channels were not seen on gross inspection of the lungs, numerous dilated vessels were apparent on histologic examination. Angiomas of the mucous membranes and the knee-joint capsule supported the diagnosis of hereditary hemorrhagic telangiectasis. Cirrhosis of the liver was present. Pulmonary fistulas may develop in patients with hepatic cirrhosis under the influence of factors similar to those resulting in the formation of cutaneous spider angiomas; severe hepatic cirrhosis occurring in a young patient may occasionally result in clinically significant pulmonary vascular proliferation.

M. J. SMALL

Funnel Chest in Adults and Children. Clinical Observations. P. TALA* and E. LAUSTELA. *Ann. chir. et gynaec. Fenniae*, Fasc. 2, 1959, 48: 177–188. (*U. of Helsinki, Finland)

A discussion is presented based on 16 cases subjected to surgery.

J. HAAPANEN

The Hamman-Rich Syndrome. Acute, Diffuse, Interstitial Fibrosis of the Lung: Pulmonary Cirrhosis (in German). F. J. SANEN.* *Wien. klin. Wchnschr.*, May 15, 1959, 71: 347–350. (*Taunton State Hosp., Mass.)

The syndrome of acute, diffuse, interstitial pulmonary fibrosis, or of acute fibrosing interstitial pneumonia, has been well described by Hamman and Rich, but had been described as early as 1898 by Rindfleisch and, again, in 1915 by Hansemann. Up till now, 52 cases of the Hamman-Rich syndrome have been published, with a slight but insignificant preponderance in the male. The age of these cases ranged from 9 to 81 years, with most of the patients 45 to 60 years old.

The physical findings are not characteristic; usually there are an accentuated pulmonic second sound, moist rales, wheezing, cyanosis, and clubbed fingers. The roentgenographic picture is not specific either: both lungs are involved. In the beginning there are increased markings due to increase of the interstitial connective tissue. Later, inhomogenous shadows are seen. Reticulate and nodular infiltrations occur. In the final state the picture may resemble that of a congested lung, of miliary tuberculosis, or of bilateral fulminant bronchopneumonia. The laboratory findings are not characteristic either. Leukocytosis and polycythemia may occur.

A definite diagnosis is possible only by biopsy or autopsy. Histologic examination reveals progressive proliferation of the interstitial connective tissue, hyperplasia of the alveolar walls, and necrosis of the alveolar and bronchiolar epithelium without significant leukocytic infiltration or exudate. Macroscopically, the hobnailed or cobblestone-like surface of the lung is characteristic. Death is usually due to cardiopulmonary insufficiency caused by alveolar-capillary block.

Therapy is symptomatic. Corticosteroids may have a beneficial effect. The etiology of this disease is not known. It may be caused by several factors, constitution or heredity apparently having some significance. Or it may be caused by some virus. A relation to the collagen diseases seems to exist.

A case history of the Hamman-Rich syndrome is presented: an 81-year-old woman died from cardiopulmonary insufficiency after having been sick for only 17 days. In this case an aplastic anemia was also present.

G. C. LEINER

Chest Deformities in Congenital Heart Disease. H. DAVIES.* *Brit. J. Dis. Chest*, April, 1959, 53: 151–158. (*National Heart Hosp., London, England)

The chest deformity associated with atrial septal defects (A. S. D.) differs clinically and roentgenographically from that of ventricular septal defects (V. S. D.). In A. S. D. there is a unilateral bulge which lies directly over the right ventricle and is considered to be directly due to the pressure of this enlarged chamber. In V.S.D. there is a bilateral upper sternal protuberance, the anteroposterior diameter of the chest is increased, and there is a tendency for the lower ribs to be drawn in on inspiration.

The "V.S.D. type" of deformity was found, on a roentgenographic basis, in ½ of 45 cases of V.S.D. in which adequate lateral chest films were available, together with cardiac catheterization data. The incidence of the V.S.D. type of deformity in cases of V.S.D. is approximately the same as that of the A.S.D. type of deformity in cases of A.S.D. In V.S.D., the deformity, which is similar to a pigeon-breast deformity, was associated in most cases with pulmonary hypertension. It is considered to be much more common when the pulmonary vascular resistance is raised than when it is not. It occurs in the "Eisenmenger syndrome" in only a minority of cases.

It is suggested that the deformity of V.S.D. may be due to changes in the mechanical properties of the lungs, and that the combination of increased pulmonary vascular resistance and increased pulmonary blood flow increases the stiffness of the lungs (after author's summary).

M. J. SMALL

Single or Double Segments of the Lung Occupying a Hemithorax. R. H. OVERHOLT,* E. A. GAENSLER, and J. A. BOUGAS. *New England J. Med.*, July 2, 1959, 261: 10–18. (*Overholt Thoracic Clinic, Boston, Mass.)

Because no data are available from the literature concerning clinical course and pulmonary function with single-segment or double-segment occupancy in a hemithorax, a late postoperative study of such patients was made. Observations included clinical status, contribution of the remaining segment or segments to total function, relative "efficiency" or the ventilation-perfusion relation of this lung tissue, and the function of these segments in terms of normal or predicted performance.

Clinical and physiologic data of this study leave no doubt that, in most cases, 1 or 2 healthy residual segments on 1 side may contribute greatly to over-all functional capacity. In patients with intact and presumably healthy opposite lungs, preservation of 1 or 2 segments resulted in greater over-all function than is ever seen after pneumonectomy. A single segment was found to contribute as much as 23 per cent, and 2 segments as much as 33 per cent to total oxygen uptake at rest. In addi-

tion, the residual lung tissue prevented or greatly reduced mediastinal shift to the side of operation.

After extensive contralateral resection, the 1 or 2 remaining segments made a larger contribution to the greatly taxed respiratory economy and, in extreme cases, were responsible for nearly half the total respiratory work. In nearly all cases with functioning residual segments, their ventilatory contribution was relatively greater than their participation in oxygen uptake. This unfavorable ventilation-perfusion ratio may have been disturbing in 1 case.

The functional contribution of the single and double segments in patients with healthy opposite lungs was greater than that predicted from theoretic consideration. In 4 cases with extensive disease or resection on the opposite side, the residual lung tissue was probably necessary for life.

M. J. SMALL

Spontaneous Hemopneumothorax in a Hemophiliac. J. F. BURKE* and E. W. SALZMAN. *J. A. M. A.*, April 4, 1959, 169: 1623–1625. (*Harvard Med. School, Boston, Mass.)

A case of spontaneous hemopneumothorax in a patient with hemophilia is described. Thoracotomy and rib resection were successfully performed with the use of fresh frozen plasma. The patient developed a *Staphylococcus pyogenes* empyema later on and died a few days after a decortication procedure.

H. ABELES

Löffler's Syndrome During a Case of Distomiasis (in French). R. GENEVRIER* and M. BOURGEOIS. *Semaine hôp. Paris*, May 4, 1959, 35: 1561–1562. (*Societé Francaise Allergic, Paris, France)

A patient who had eaten cress in an area where sheep are often infected with parasites was diagnosed as having distoma by means of a complement-fixation test. The lungs showed a hypereosinophilic infiltrate. The authors discuss fluke worm infection in the etiology of Löffler's syndrome and isolated pulmonary distomiasis.

E. LYON

Traumatic Mediastinal Hematoma: Report of Unusual Case. C. COTE,* C. H. HODGSON, and F. H. ELLIS, JR. *Proc. Staff Meet. Mayo Clin.*, May 13, 1959, 34: 264–268. (Mayo Clinic, Rochester, Minn.)

A case of traumatic mediastinal hematoma occurring in a 45-year-old man as the result of an automobile accident is described. Only 3 similar cases in which there was evidence of demonstrable injury to adjacent structures have been reported. The diagnosis was made by exploratory thoracotomy since chest roentgenograms showed progressive mediastinal enlargement suggesting tumor or aneurysm.

E. A. RILEY

Cavitation in Pulmonary Sarcomatous Metastases with Hypertrophic Osteoarthropathy. P. E. BALDRY.* *Brit. J. Dis. Chest*, April, 1959, 53: 173–176. (*Ashford Hosp., London, England)

Cavitation occurs rarely in multiple metastatic deposits in the lungs. A case is reported of a man who developed cavities in multiple lung nodules. The correct diagnosis was difficult because, in addition to the fact that 2 years previously he had had a sarcoma removed from a thigh muscle, he also had been in contact with his daughter who suffered from pulmonary tuberculosis and who had intermittently positive sputum for several years. A further unusual feature of this case was the development during the progress of the disease of hypertrophic pulmonary osteoarthropathy.

M. J. SMALL

Pulmonary Histoplasmosis in Africa with Diffuse Suppuration Appearing after a Year of Evolutive Course (in French). A. P. JARNIOU,* G. KERBRAT, A. MOREAU, and DUVAL. *Presse méd.*, April 22, 1959, 20: 824–827. (*Pasteur Institute, Paris, France)

A case of pulmonary histoplasmosis was observed and studied in Africa. This disease has occurred predominantly in America and has been very rare on the African continent. It presented a difficult diagnostic problem because of its rarity and because there were no clinical manifestations in the beginning. Only the chest films showed abnormal findings, and the disease developed slowly through a period of one year to a bronchopulmonary suppuration which responded favorably to treatment with amphotericin. The authors consider this drug the drug of choice in the treatment of histoplasmosis, for which no effective treatment was known up till now. The case was especially studied in reference to its mycologic aspects.

E. LYON

Sarcoidosis: Report of Three Cases in Siblings under Fifteen Years of Age. E. L. KENDIG, JR.,* R. L. PEACOCK, and S. RYBURN. *New England J. Med.*, May 7, 1959, 260: 962–966. (*Med. College of Virginia, Richmond)

Three cases of sarcoidosis in siblings under 15 years of age are reported. The patients described live in a predominantly rural area in the southeastern part of the United States where there are pine forests and sandy soil and the chief crop is peanuts. Neither contact with a known infectious agent nor unusual dietary regimen can be elicited from the history.

Indeed, the only significant feature is evidence available to the local health department that, in this county, (Nansemond County, Virginia) with an estimated population of 38,000, there are 27 cases of proved or probable sarcoidosis. It appears that a more thorough investigation of this area is indicated. It is said that practically all of the persons in this locality known to be affected have worked with peanuts, either in the fields or in factories, and the possibility that in this particular area the causative agent is disseminated in the dust from peanut shells must be considered.

M. J. SMALL

Clinical and Pathological Studies of Joint Involvement in Sarcoidosis. L. SOKOLOFF* and J. J. BUNIM. *New England J. Med.*, April 23, 1959, 260: 841–847. (*Nat. Inst. of Arthritis and Metabolic Diseases, U. S. Dept. of Health, Education, and Welfare, Washington, D. C.)

Five cases of sarcoidosis are presented in which polyarthritis was a conspicuous feature. In each there were extra-articular manifestations of sarcoidosis. Biopsy established the presence of noncaseating tubercles in the synovial tissue in 3, and a submiliary granuloma suggestive of sarcoid in a fourth patient. In the fifth the synovitis was of nonspecific histologic nature. No fungi or acid-fast organisms could be recovered from the lesions. The joint disease was not secondary to that in the bone and must be regarded as an intrinsic manifestation of the sarcoidosis. There is reason to believe that joint involvement of this sort is more common than has been recognized previously, and that it has been mistakenly diagnosed as rheumatic fever or rheumatoid arthritis. The frequency with which the lesion is associated with erythema nodosum invites speculation about the role of hypersensitivity in the pathogenesis of the disorder, and also raises the question whether the arthritis and adenopathy of erythema nodosum are not themselves inherently related to sarcoidosis. Synovial biopsy may be of diagnostic value in such cases (Authors' summary).

M. J. SMALL

Clinical and Roentgenographic Improvement of Pulmonary Sarcoidosis Treated with Prednisone (in Italian). L. DONNO,* P. D'ANGELO, and N. PATRONI. *Ann. med. Sondalo*, March–April, 1959, 7: 71–90. (*V. Sanatoriale, Sondalo, Italy)

The clinical history and laboratory data are presented on 2 male patients, 30 and 21 years old, with sarcoidosis (the diagnoses were not histologically substantiated) with lymph node- and pulmonary localization. Prednisone was given daily in a dosage of 20 to 30 mg., alternating with 50 units of corticotropin for 2 days after an interval of 13 to 28 days. Treatment lasted for 6 to 7 months without untoward symptoms. The results were good and improvement was consistent.

I. ARCHETTI

The Acute Pneumopathies of Occupational Origin (in French). M. MARCHAND. *Lille méd.*, March, 1959, 3: 134–149.

Most of the occupational lung diseases are chronic in nature. However, the author feels that acute pneumopathies of occupational origin are much more frequent than is usually realized. He gives a comprehensive review of these acute conditions. They are described in detail, and grouped according to the causative agent as follows: chemicals which can be inhaled (42 such chemicals are named); metallic fumes inhaled in the process of welding; substances inhaled while filling a silo with fodder and grains; various insecticides, also carbonoxides and hydrocarbons, lipoids and oils. Certain metals which can be inhaled receive special attention, such as beryllium, manganese, and osmium. Infectious pneumopathies due to occupational hazards are described, most of them well known, and also those pneumopathies due to parasites.

E. LYON

Diffuse Pulmonary Schistcsomiasis. Report of Two Cases Proved by Lung Biopsy. J. H. RICHERT* and R. B. KRAKAUR. *J. A. M. A.*, March 21, 1959, 169: 1302–1306. (*Fitzsimons Army Hosp., Denver, Colo.)

Two male Puerto Rican soldiers were hospitalized because of signs and symptoms of chronic pulmonary disease. In one, a lung biopsy specimen obtained at exploratory thoracotomy was found to contain lesions characteristic of pulmonary schistosomiasis, and ova of *Schistosoma mansoni* were identified. In the other, the ova of *S. mansoni* were first found in the feces; later the characteristic ova were also found in lung tissue obtained by wedge biopsy. Both patients responded quite well to treatment with stibophen (Fuadin®).

H. ABELES

"Diagnostic Small Thoracotomy" in Idiopathic Pleural Effusion. F. T. LANSDEN* and W. H. FALOR. *J. A. M. A.*, July 18, 1959, 170: 1375–1379. (*Akron City Hosp., Ohio)

A method of small thoracotomy with pleural biopsy was used for 17 cases of pleural effusion after all other medical and laboratory procedures had failed to establish a diagnosis. The thoracic cavity was entered by an incision not exceeding 20 cm. in length, thus permitting thorough ex-

ploration of the thoracic cavity and removal of biopsy specimens.

H. ABELES

Bronchial Adenomas. O. CH. POCK-STEEN.* *Acta radiol.*, April, 1959, 51: 266–272. (*Rigshospitalet, Copenhagen, Denmark)

Nineteen patients treated for bronchial adenomas over the past 15 years are reviewed. Five occurred in men, and 14 in women. The youngest was 17 and the oldest 69 years of age. Seventeen patients were symptomatic with attacks of fever, recurrent pneumonia, productive cough, and fatigue. Hemoptysis occurred in 50 per cent of the cases. Chest pain and dyspnea were less common.

Ordinary chest films, bronchography, and tomography were used in the diagnosis. Four of the tumors were peripheral. Fifteen were juxta-hilar and, of these, 12 presented evidence of massive atelectasis. Other roentgen findings were bronchiectasis and abscess cavities; 1 had a spontaneous pneumothorax. The lesion had a tendency to occur most frequently in the right upper lobe. More than ½ of the cases presented a roundish lesion in the bronchial branch when studied by tomography and bronchography. Lobectomy and pneumonectomy were the prevalent methods of treatment, although local excision was employed in 3 of the cases.

The pathology is discussed and the tumors are classified into cylindromas, carcinoids, and a third group referred to as mucous-producing. The cylindroma was the only type encountered in the central type of case, and the author considers this group the most likely to undergo malignant changes. Invasion characteristics were found in 9 of the 19 cases. Although a rare tumor, bronchial adenoma is the most common of the benign types. A review of the literature reveals an average period of 12–15 years before regional lymph node metastasis. A differential diagnosis is given.

E. SHABART

Aspergilloma Superimposed upon an Apparently Inactive Cavity (in French). A. BERNOU,* J. ROUSSELOT, R. GOYER, L. MARÉCAUX, and J. TRICOIRE. *Rev. tuberc., Paris,* April, 1959, 23: 440–448. (*Chateaubriant, France)

A case report is presented of a female patient with a large tuberculous cavity in the left upper lobe and positive sputum who was treated with isoniazid and PAS. There was reversal of infectiousness, and the cavity assumed a bullous appearance which remained unchanged for 4 years. The patient then developed recurrent hemoptyses; roentgenograms showed marked shrinkage of the cavity with irregular thickening of its walls and a mass protruding into the lumen from its inferior border. Pulmonary resection was performed. Histologic examination showed an aspergilloma within a bullous cavity, the walls of which showed no residual tuberculous foci.

V. LEITES

Malignant Mesothelioma of the Pleura. K. SIROLA.* *Ann. chir. et gynaec. Fenniae,* Fasc. 2, 1959, 48: 226–233. (*U. of Helsinki, Finland)

Two cases of malignant pleural mesothelioma are reported. The tumors were found in males aged 41 and 56 years. The first symptom was pleurisy with effusion. In one case, the localized tumor was removed by lobectomy. In the other case, only an exploratory thoracotomy was possible because of the diffuse spread of the tumor. The invasion of the neoplasm continued despite radiation therapy, and both patients died, one 6 months, and the other, 18 months after the operation.

J. HAAPANEN

Cytologic Diagnosis of Pulmonary Neoplastic Processes (in Italian). A. FERRARIS,* E. D'AMORE, and G. CHIODI. *Gior. ital. tuberc.,* January-February, 1959, 13: 3–10. (*U. of Genoa, Italy)

The data in this paper on cytologic diagnosis of pulmonary neoplastic processes are based upon 372 microscopic examinations. The technique of choosing the material—and the importance of this factor should be particularly emphasized—the methods of staining (Papanicolaou and May-Grünwald-Giemsa), the criteria followed in diagnosing the malignancy (which are divided into 2 groups, principals and secondaries) are briefly discussed.

The value of the cytologic examination as a method of choice for an early diagnosis, particularly when the others are negative, is recognized. The use of this method is also very important when the pathologic processes are too peripheral so that they cannot be reached by bronchoscopy.

I. ARCHETTI

Association of Type 2 Hemadsorption (Parainfluenza 1) Virus and Asian Influenza A Virus with Infectious Croup. A. VARGOSKO,* R. M. CHANOCK, R. J. HUEBNER, A. H. LUCKEY, H. W. KIM, C. CUMMING, and R. H. PARROTT. *New England J. Med.,* July 2, 1959, 261: 1–9. (*Children's Hosp., Washington, D. C.)

Croup is one of the most dramatic, frightening, and dangerous syndromes in pediatrics. In some cases, when treatment is not available, death results. In the fall of 1957, a study to isolate new viruses and to associate these agents with clinical illness, including croup, was initiated by the Virus Section, Research Foundation of the Children's

Hospital of the District of Columbia, and the Laboratory of Infectious Diseases, National Institutes of Health.

Evidence for a viral infection was detected in 64 per cent of 47 patients with infectious croup studied over a 1-year period in the Washington, D. C., area. Evidence for infection with Type 2 hemadsorption virus was found in 36 per cent of all patients with croup, and this virus was associated with croup throughout the year. The virus of Asian influenza alone was associated with 36 per cent of patients with croup studied during the Asian influenza epidemic, and 19 per cent through the year. Nine per cent of croup illness was associated with adenovirus, hemadsorption virus Type 1, or croup-associated virus. Infection with Type 2 hemadsorption virus was found predominantly in boys, but its clinical features were not distinctive.

M. J. SMALL

The Training of the Greatest of Chest Physicians: Laennec—Part II: Paris. E. A. UNDERWOOD.* *Brit. J. Dis. Chest*, April, 1959, 53: 121–124. (*Wellcome Historical Museum & Library, England)

TUBERCULOSIS, PULMONARY

Medical Progress: Tuberculosis. T. L. BADGER.* *New England J. Med.*, July 2, 1959, 261: 30–36; July 9, 1959, 261: 74–81; July 16, 1959, 261: 131–139. (*Harvard Med. School, Boston, Mass.)

This comprehensive and well-documented review of recent progress in our knowledge of tuberculosis covers the following topics: epidemiology, case finding, the tuberculin test as a case-finding technique, the types of tuberculin tests, diagnosis, roentgenographic study, case-finding summary, the roentgen examination as a case-finding technique, medical treatment of tuberculosis, rest versus ambulation as a basic plan of treatment, chemotherapy, the target point, isoniazid alone, other drugs, re-treatment cases, home-care treatment, relapse, corticosteroids in treatment, chemoprophylaxis, BCG vaccination, pleurisy with effusion, genitourinary tuberculosis, meningeal tuberculosis, miliary tuberculosis, surgery in tuberculosis, selection of preoperative drug regimens, type of surgical procedures, pulmonary function in the surgery of tuberculosis, and the future of tuberculosis.

M. J. SMALL

The Mental Image of Tuberculosis. Results of a Questionnaire (in French). P. FRÉOUR,* M. SERISE, P. COUDRAY, and G. MALÉSIEUX. *Rev. tuberc., Paris*, April, 1959, 23: 428–439. (*Bordeaux, France)

A questionnaire was sent to 200 patients with pulmonary tuberculosis in order to determine the present-day concepts patients have of their disease.

From the first onset of symptoms until the establishment of diagnosis of tuberculosis, 82 of 100 patients did not think of tuberculosis as a cause of their complaints; only 18 feared this possibility. Of 10 patients with hemoptysis, only 3 were worried about having tuberculosis. This attitude is very much in contrast to the past, when anxiety concerning tuberculosis was widespread. Today tuberculosis is believed to be benign, and does not represent a major health concern. Sixty per cent of the patients hoped to achieve cure of tuberculosis within 4–6 months. (Of patients who were sick for more than 2 years, only 44 per cent expected cure within this time.) In spite of this optimistic view, 51 per cent expected to have a relapse. A total of 74 per cent believed that tuberculosis can be either cured or stabilized.

Sixty per cent believed that tuberculosis was a contagious disease; 16 patients did not consider it contagious; 24 did not know. However, of 62 patients whose sputum was positive and who had been informed of this, only 13 (1 of 5) believed that they themselves were contagious. Of 200 patients, only 46 per cent said they were observing or going to observe precautions toward their children. Sixty-eight per cent believed that patients with a history of active tuberculosis could marry and have children; 23 per cent did not know; 9 per cent answered this question with "no."

The tendency to minimize the gravity of tuberculosis was less evident in answers regarding the ability to resume work. Of 100 patients, 43 did not believe they could resume their former jobs; 27 did not know; 30 thought they could go back to their former occupation; 61 per cent believed that tuberculosis was a definite handicap to manual labor. The wish to change their occupation was expressed by 63 per cent of laborers, 33 per cent of seamen, 30 per cent of workers, and 28 per cent of farmers.

Confidence in their physicians was expressed by 85 per cent of patients; however, only 60 per cent thought that the primary concern of doctors is the cure of their patients; 23 per cent believed that financial interests were as important. Sixty per cent thought that doctors are interested in patients as human beings; 28 per cent did not know; 12 per cent answered "no." Thus, while the patients on the whole showed great confidence in medical science regarding eradication of tuberculosis, the answers indicate a certain malaise in the patient-doctor relationship.

V. LEITES

Tumoral Unilateral Silicosis and Atypical Acid-fast Chromogenic Bacilli (in French). G. Despierres,* P. Galy, J. Viallier, and J. Vauzelle. *Rev. tuberc., Paris*, February-March, 1959, 23: 204–213. (*Lyon, France)

A case report of nodular silicosis with a dense excavated mass at the right apex is presented. Sputum, pleural fluid of a terminal empyema, and pus of the pulmonary cavity showed the presence of acid-fast mycobacteria which were chromogenic and nonpathogenic for laboratory animals, although they seemed to have caused the fatal outcome. The anatomic, therapeutic, medicolegal, and social problems raised by these findings are discussed.

V. Leites

Reversal of Tuberculin Reaction in Early Tuberculosis. J. M. Adams,* V. A. Kalajan, B. O. Mork, M. Rosenblatt, W. J. Rothrock, and B. J. O'Loughlin. *Dis. Chest*, April, 1959, 35: 348–356. (*U. of California Med. School, Berkeley)

The traditional concept that a positive tuberculin reaction will remain so for the rest of the person's life has been widely accepted for many years. The present study was carried out to detect tuberculosis in its earliest phases by means of a positive tuberculin test in individuals who reacted negatively to previous test or tests.

In the past 2 years, 160 individuals have been discovered who had known negative reactions which converted to positive within 3 months, or within 12 months in infants under 1 year of age. Of 121 individuals tested two or more times, 68 have changed from a positive reaction to a negative one. The rate of reversal was 76 per cent when the test was 1+, 50 per cent when 2+, 38 per cent when 3+, and only 1 individual reversed who was 4+. Those with negative chest roentgenograms showed a high rate of reversal; no change in skin reactivity was found when the findings indicated a "presumably active" lesion. This study emphasizes the value of repeating the skin test.

E. A. Rouff

The Alterations of Peripheral Bronchi in Early Pulmonary Tuberculosis Studied by Bronchography (in Italian). G. De Michele,* G. Melillo, and G. Mauro. *Arch. tisiol.*, March, 1959, 14: 245–256. (*U. of Naples, Italy)

The modifications of the peripheral bronchi in early pulmonary tuberculosis, both in its primary and postprimary forms, were studied using bronchography. The variations were found to be manifold (occlusion of the bronchial branches, ectasia, bead-like images, distortion, et cetera) and not characteristic of the tuberculous process. Alterations were generally strictly related to the diseased parenchymal section of the lung, and influenced by the anatomic substratum of the process and by the length of illness. Chronicity was found to favor the progressiveness of bronchial alterations and, consequently, the origin of bronchiectases.

I. Archetti

Tuberculous Pulmonary Cavities in Children (in Finnish). K. Mäkipaja* and J. N. Mäkipaja. *Duodecim*, May, 1959, 75: 378–387. (*Kinkomaa, Finland)

A total of 32 cavities was found in 20 children whose ages varied between 6 and 14 years at the onset of the disease. Four of the cavities were thick walled. In 8 cases, the disease was bilateral. All 20 patients were treated medically, usually with a combination of streptomycin, isoniazid, and PAS, for a period of 4 to 30 months. Pneumothorax was started on 2 patients, but discontinued due to adhesions after 1 and 5 months, respectively. A segmental resection was performed in 2 cases in which the cavity reopened after an initial closure. All of the patients were on bed rest for several months. A total of 21 cavities healed by cicatrization, and 2 by filling. Three patients were still under treatment. One case terminated fatally.

It is recommended that cavitary pulmonary tuberculosis before and during puberty should be treated medically for at least 18 months before contemplating any surgical intervention.

J. Haapanen

C-Reactive Protein and Katz's Index in Pulmonary Tuberculosis (in Italian). M. A. Sisti,* V. Pellegrino, and N. Sozio. *Lotta contro tuberc.*, November–December, 1958, 28: 1191–1196. (*Osp. Cotugno, Bari, Italy)

The C-reactive protein and blood sedimentation rate were studied in 180 patients with pulmonary tuberculosis. From comparison of the results, it was concluded that: the protein was absent in a high percentage of the cases (63 per cent), and also when the Katz index was high; when the protein was present there was no correlation between it and the blood sedimentation rate values. A correspondence was found in 5 cases; 4 had complications of the tuberculous process, 1 which was particularly severe.

I. Arghetti

A Comparison of Two Methods of Sputum Collection in the Diagnosis of Pulmonary Tuberculosis. R. H. Andrews* and S. Radhakrishna. *Tubercle*, June, 1959, 40: 155–162. (*Tuberculosis Chemotherapy Center, Madras, India)

It is generally accepted that a specimen of sputum collected over a period of hours, whether

an overnight specimen or a 24-hour specimen, is more valuable for the identification of *M. tuberculosis* than is one produced immediately upon request. In less favorable circumstances, such as may be encountered in diagnostic and survey work under field conditions, it may only be practicable to obtain an immediate specimen in the clinic or home. The present report gives the results of a comparison of these two types of specimens collected from 348 patients at the Tuberculosis Chemotherapy Center, Madras, using the results of direct examination by fluorescence microscopy and of culture on Löwenstein-Jensen medium.

While inferior to a "collection" specimen, a "spot" specimen can yield useful information, especially if facilities are available for culture, and from a single spot specimen the presence of *M. tuberculosis* was demonstrated by direct examination in 66.1 per cent, and by culture in 90.5 per cent of patients known to have positive sputum. Its simplicity is a major advantage for diagnostic and survey work in field conditions. Direct examination of two spot specimens gave 76.4 per cent positive results, and of two collection specimens, 83.0 per cent of positive results, so that a high proportion of cases presenting symptoms and having a roentgenographic abnormality can be confirmed as tuberculous infections without recourse to culture. The majority of the sputum specimens examined were from patients with extensive and cavitated disease whose sputum was highly positive. A comparison of the results of spot and collection specimens restricted to patients with apparently uncavitated disease confined to one or two lung zones, and from patients receiving chemotherapy whose sputum was of a lesser grading of positivity, showed a greater advantage in collection specimens.

M. J. SMALL

The Use of Steroids in Tuberculous Patients with Untoward Reactions to Anti-Microbial Therapy. R. GROSSMAN. *Dis. Chest*, June, 1959, 35: 615–624.

The combined use of steroids and antimicrobial agents appears to have produced a prompt salutary effect in extinguishing simultaneously drug reaction (pruritic rash, angioedema, fever, and hepatitis) and disease toxicity. There was regression of tuberculous exudative and recent infiltrative lesions. The cases have now been observed from 21 to 26 months subsequent to steroid therapy; no adverse effect on the pulmonary disease has been noted to date.

E. A. ROUFF

A Study of Re-treatment Cases vs. Original Treatment Cases of Pulmonary Tuberculosis. W. J. STEININGER* and W. L. HOWARD. *Dis. Chest*, May, 1959, 35: 480–493. (*Wm. H. Maybury Sanatorium, Northville, Mich.)

A primary problem in the current treatment of tuberculosis is the ever-increasing number of patients who have had interrupted periods of antimicrobial therapy. Over a 2-year-period (January, 1955–December, 1956) 56.5 per cent of admissions to Maybury Sanatorium (643 cases) were original treatment cases, and the remainder (495 cases) repeat cases. Significant differences in background and treatment response were noted in the two groups. The typical re-treatment patient was unskilled, alcoholic, with far advanced fibrocavitary disease of 2 years duration, and an abscondee with 1 or 2 periods of previous therapy. The re-treatment patient lagged far behind in roentgenographic clearing, cavity closure, and sputum conversion; he experienced more roentgenographic worsening, bacillary resistance, and death from tuberculosis.

E. A. ROUFF

Pulmonary Tuberculosis Due to Primary Infection with Streptomycin-resistant Tubercle Bacilli (in Japanese). T. YASUTAKE,* J. NAGAKI, T. KOJIMA, M. SHIRABE, and T. MATSUZAKI. *Clin. Resp. Organs*, July, 1959, 14: 563–565. (*Kumamoto U. School of Med., Kumamoto, Japan)

A case report is made of pulmonary tuberculosis in a nurse in whom tubercle bacilli in the sputum before treatment were resistant to 100 γ of streptomycin per ml., and susceptible to 1.0 γ of PAS per ml. and 0.1 γ of isoniazid per ml., respectively. Her tuberculin reaction was negative and she was vaccinated with BCG in 1949. She became positive for tubercle bacilli in 1950 and remained positive throughout the years 1951 to 1953, undergoing a skin test each year. She was transferred to the Service of Tuberculosis Wards *after* her tuberculin reaction turned positive. Her course was uneventful with treatment with PAS and isoniazid.

The positive conversion of her tuberculin reaction might have been due to primary infection with streptomycin-resistant tubercle bacilli instead of due to BCG; or, the secondary infection with the former might have occurred some time after vaccination with BCG.

I. TATENO

Meticorten as Adjuvant Therapy in Chronic Pulmonary Tuberculosis with Emphysema. I. G. EPSTEIN,* P. K. BORNSTEIN, K. G. S. NAIR, and B. CARPIO. *Dis. Chest*, May, 1959, 35: 509–516. (*New York Med. College, N. Y.)

This study involved 44 patients with chronic active pulmonary tuberculosis and emphysema.

All had received at least 2 years of chemotherapy with no clinical or roentgenographic improvement. Half of the patients were used as controls. All received antituberculosis drugs as determined by susceptibility studies. Twenty-two received, in addition, 20 mg. of prednisone daily in 4 divided doses for an average of 5.5 months. The untoward effects noted included diabetes in 3 patients and cardiac decompensation in 4 others. No significant roentgen deterioration was noted. Ten of the patients who had had repeated hemoptyses were completely free of bleeding. After 24 weeks, half of the treated patients had tubercle bacilli in their sputum.

E. A. ROUFF

Clinical Significance and Prognostic Value of C-reactive Protein in Pleuropulmonary Tuberculosis (in Italian). F. MONCALVO* and G. PIAZZA. *Ann. med. Sondalo*, March–April, 1959, 7: 91–103. (*U. of Pavia, Italy)

The value of C-reactive protein was compared with the blood sedimentation rate in 97 patients with pleuropulmonary tuberculosis of varying severity and at different stages of evolution. Samples of serum were taken at the beginning of the study and after an interval of 3 to 7 months. The C-reactive protein test was also done in the serum and pleural effusion fluid of 11 other patients.

The main observations were that C-reactive protein was present in a very high percentage of cases; there was no constant correspondence between increased blood sedimentation rate and the presence of C-reactive protein. It was concluded that the C-reactive protein test was useful for its prognostic value, particularly in cases near resolution. C-reactive protein was found in 10 of the 11 cases with pleural effusions, both in the serum and in the fluid.

I. ARCHETTI

TUBERCULOSIS, NONPULMONARY

Two Cases of BCG Adenitis (in Turkish). C. GURSON,* B. PARS, S. ERSEK, and M. BERKER. *Tuberkuloz*, January, 1959, 12: 34–38. (*Istanbul, Turkey)

Of 7,791 BCG-vaccinated persons, 0.022 per cent developed the complication of adenitis. In 2 cases studied, it was found that BCG adenitis does not drain to the surface, but becomes hard and calcified in 2 to 4 years. Infected lymph nodes revealed histopathologic characteristics of primary hyalinization and calcification along with occasional localized specific granulation tissue.

S. SOLU

Splenectomy in Splenomegalic Forms of Boeck's Disease (in French). J. TAPIE,* J. MONNIER, P. FERRET, Y. LE TALLEC, and MLLE BOURDIN. *Presse méd.*, April 22, 1959, 67: 805–808. (*Toulouse, France)

Splenectomy is considered the most efficient therapy in cases of splenomegaly caused by Boeck's disease. The operation is indicated when the splenomegaly becomes chronic and is complicated by the emergence of "hypersplenism," that is, anemia and various pathologic changes in the blood. Treatment with cortisone is indicated over a long period of time, as well as treatment for whatever changes the blood presents. The authors describe a case which they have observed personally, and other cases in the literature. Lung changes suggestive of sarcoidosis were also present in these cases, but caused no symptoms.

E. LYON

On the Increasing Incidence of Benign Lymphogranulomatosis. Clinical Evaluation of Thirty Cases (in German). R. SCHMUTZLER.* *Ztschr. Tuberk.*, April, 1959, 113: 19–31. (*Universitätsklinik, Rostock, Germany)

Thirty cases of benign lymphogranulomatosis are described. The incidence was greater in females (17 cases), and greatest in the age groups of 20 to 29 and of 40 to 49 years. The lungs were involved in 29 instances, the lymph nodes in 12, the skin in 8, the eyes in 6, the salivary and lacrimal glands in 3, the nerves in 2, the mucous membranes in 2, and the bones in only one instance. The diagnosis was verified in 12 patients by histologic examination, and in the other 18 by history, pathogenesis, and biochemical data. In 20 per cent of the patients the tuberculin skin test was clearly positive. Hypercalcemia was present in ⅔ of all cases, with a maximum of 14.2 mg. per 100 ml. Hypergammaglobulinemia (25.7 per cent) was discovered by electrophoresis. Blood count showed slight monocytosis, and eosinophilia of 4–5 per cent.

Fifteen patients had no specific treatment. Ten patients were treated alternatively with cortisone and corticotropin for 4 months. The end result was essentially the same in treated and untreated cases, although healing was hastened by the treatment.

J. HAAPANEN

Scrofula: Then and Now—1500 Years of Tuberculous Lymphadenitis in Accessible Nodes. C. W. LESTER.* *Arch. Pediat.*, May, 1959, 76: 189–195. (*70 E. 80th St., N. Y. 21, N. Y.)

Scrofula, i.e., tuberculous lymphadenitis in the neck, axilla, and groin, has been in the public consciousness for at least 1,500 years. It has been subjected to many forms of treatment, among

which the "king's touch" was in vogue most of the time. In judging the worth of treatment, it must be remembered that spontaneous improvement and cure can take place in due course. Treatment depends upon the presence or absence of other tuberculous lesions, and in all cases it should be initiated by general hygienic measures and chemotherapy.

Surgery is an effective and safe form of treatment. It implies the removal of all of the diseased nodes with the preservation of all of the important structures, especially the eleventh cranial nerve. The incisions are made in or parallel to natural creases in the neck so that when they heal the scars will not be noticeable. It should be employed when chemotherapy has not produced an adequate result, and not unduly delayed in the absence of tuberculosis in other organs.

M. J. SMALL

Pancytopenia Associated with Disseminated Tuberculosis. W. COOPER.* *Ann. Int. Med.*, June, 1959, 50: 1497–1501. (*U. of Witwatersrand, Johannesburg, South Africa)

A case is described in which pancytopenia with splenomegaly were associated with high fever, sweating, and loss of weight. The patient was treated empirically with streptomycin and isoniazid because the pronounced constitutional symptoms suggested underlying tuberculosis. Pancytopenia associated with splenomegaly and a marrow of normal cellularity suggested that hypersplenism was responsible for the blood changes. Although splenectomy is not often recommended in secondary hypersplenism, satisfactory results have been achieved in a number of conditions, as in Gaucher's disease and in kala-azar. In the case described here, chemotherapy did not bring about the expected improvement, and the patient appeared to be resistant to such treatment. The prompt and satisfactory improvement in the blood picture following removal of the spleen proved that splenectomy was a justifiable procedure.

T. H. NOEHREN

Use of Isoniazid and PAS in the Treatment of Genitourinary Tuberculosis (in French). J. CIBERT and J. BARBIER. *Presse méd.*, April 25, 1959, 21: 841–842.

Isoniazid combined with PAS have been used for the past 2 years for the treatment of 44 patients with genitourinary tuberculosis. Thirty patients had renal tuberculosis, and 44, tuberculosis of the male genital organs. The results were favorable and the treatment seems to be very effective. There was only 1 failure in this series, and 4 cases of intolerance toward the drugs. In renal tuberculosis, the combination of streptomycin and isoniazid–PAS seems to be as effective as, or even superior to, any other current treatment.

E. LYON

Low Back Pain and Tuberculosis. G. BAKALIM.* *Ann. chir. et gynaec. Fenniae*, Fasc. 2, 1959, 48: 141–150. (*U. of Helsinki, Finland)

During the years 1951–1953, tuberculous spondylitis was diagnosed in 8.3 per cent of 3,203 cases of low back pain. The importance of tomography and the sedimentation rate in the diagnosis of tuberculous spondylitis is stressed.

J. HAAPANEN

Gross Anatomic and Histologic Aspects of Renal Tuberculosis Treated with Antimicrobials (in Italian). M. PEZZA.* *Arch. tisiol.*, December, 1958, 13: 1152–1176. (*U. of Naples, Italy)

Data are presented based upon the study of the kidneys of 11 patients with renal tuberculosis who were first treated with antimicrobials and subsequently by nephrectomy. The treatment with antimicrobials changed the anatomic and histologic characteristics of the renal tuberculosis. Emphasis was particularly placed on observation of cavities with clean walls and the absence in many patients of caseous processes. Instead, thick connective bundles and granulation tissue were observed, as well as histiocytic elements with epithelioid and giant cells.

I. ARCHETTI

THORACIC SURGERY

Compliance in Apneic Anesthetized Adults. P. SAFAR* and L. AGUTO-ESCARRAGA. *Anesthesiology*, May-June, 1959, 20: 283–289. (*Baltimore City Hosp., Maryland)

Fifty-four patients without pulmonary disease were studied to show that compliance of lungs plus thorax is a function of the position of the patient on the operating table, of hypo- or hyperventilation, and of the degree of muscle relaxation. Even when the above factors are maintained constant, there is individual variation of the lung-thorax compliance. The method is described in detail. Apnea is produced by a variety of anesthetic agents plus passive hyperventilation. Compliance was found to vary from patient to patient, and the range of compliance is explained by differences in weight and body configuration. Changes of the depth of cyclopropane anesthesia did not appreciably alter compliance, providing lung inflations remained constant. Poor inflation decreased, while over-inflation increased the values of compliance. *d*-Tubocurarine and succinylcholine injected during cyclopropane anesthesia increased

compliance slightly. Position changes on the operating table altered compliance from 60–120 per cent of the supine control values. The prone position and Trendelenburg position decreased compliance considerably.

E. SHABART

Pulmonary Atelectasis following Lobectomy (in Japanese). M. SHIOZAWA,* S. TAJIRI, E. HORIE, H. YASUNO, I. KINOSHITA, and T. NAKANO. *Jap. J. Clin. Tuberc.*, July, 1959, 18: 472–482. (*Institute of Tuberculosis Prevention Society, Tokyo, Japan)

Among 152 lobectomies and 116 segmental lobectomies performed during a period of 2 years, 7 cases (4.1 per cent) of pulmonary atelectasis followed. Four of the patients were males, and 3 females. The atelectasis occurred in the same side as the operated lung in 6 cases and in the opposite lung in 1 case. Preoperative vital capacity was within plus or minus 20 per cent of expected values in all of the cases, and there were no "wet" cases. Atelectasis occurred in the right middle lobe in 5 cases, in the right upper lobe in 1, and in the total left lung in 1. It developed within 2 days of operation in 5, and after 1 month in 2. It disappeared within 4 days in 4 without any specific treatment, and lasted for more than 5 months, necessitating operation, in 3. Review of the literature and analysis of the etiologic factors, treatment, and prevention of the disease are discussed.

I. TATENO

The Bronchial Spread of Lung Cancer. R. E. COTTON.* *Brit. J. Dis. Chest*, April, 1959, 53: 142–150. (*Middlesex Hosp., London, England)

A series of 100 surgically resected specimens of primary lung cancer were examined in detail to determine the methods and extent of spread in relation to the bronchus.

Direct extension in the bronchial wall proximal to the main tumor mass occurred in 12 cases. Direct involvement of other bronchi by direct extension or tumor-invaded lymph nodes is rarely seen at a stage when the tumor is still operable, with the exception of tumors close to, or involving, the major lobar orifices. Twenty-two of 32 of such tumors in this series showed distal involvement of the other lobar bronchus by parabronchial spread. In 6 cases there was invasion of submucosal lymphatics, in 4 of these it was of considerable extent, the furthest more than 1 inch. All of these cases had extensive mediastinal lymph node invasion. Extension of the tumor along the bronchial lumen by polypoid processes without further attachment to the wall was seen on 16 occasions. Thirty-two cases showed epithelial metaplasia in the bronchus proximal to the tumor; in all but 1 this was of a squamous nature. In 17 cases this was confined to the immediate proximity of the tumor; in the remaining 15 the distance varied from ¼ to 2 inches.

The practical implications of these findings are discussed in relation to the surgery of lung cancer. If these factors are kept in mind and certain criteria adhered to, there is no contraindication to lobectomy from the bronchial spread aspect. When the tumor is close to the major lobar orifices, sleeve resection of the other bronchus should be performed when a pneumonectomy is contraindicated.

M. J. SMALL

Survival in Lung Cancer. K. R. BOUCOT,* U. HORIE, and M. J. SOKOLOFF. *New England J. Med.*, April 9, 1959, 260: 742–746. (*Woman's Med. College of Pa., Philadelphia)

Data are presented on 250 consecutive proved cases of primary bronchogenic carcinoma from two official Philadelphia survey units. The 5-year survival rate, calculated from the date of the first abnormal film, was 17 per cent. There is widespread discouragement about the survival of patients with lung cancer. Surgeons have never achieved better than 10 per cent 5-year survival rates, probably because of the late stage of the disease by the time of diagnosis. However, there is evidence that efforts to detect cancer early are rewarding. The 17 per cent figure in this study is dated from the first roentgenographic abnormality at the cancer site. The prognosis was better for those without symptoms, for those with squamous-cell carcinoma, and for those whose tumors originated as solitary nodules.

In view of the currect hysteria about radiation, it is extremely important to stress the effectiveness of periodic roentgenographic study of the chest for asymptomatic persons in the cancer age group. Pending the availability of cancerocidal agents, enhanced efforts to detect early lung cancer are of the utmost importance. These include semiannual chest films of asymptomatic older persons, and exhaustive clinical study of persons in the cancer age group who have respiratory symptoms. Meticulous prolonged follow-up study should be directed toward older persons with pneumonias even if response to antimicrobial therapy has been prompt. Roentgenologists should over-read any roentgen abnormalities in the chest films of older people and, in their presence, cancer should be suspected unless prompt proof is forthcoming that the lesions are benign.

M. J. SMALL

Study of the Pulmonary Hemodynamics in a Silicotic Patient after Pneumectomy (in Italian). M. Magistretti,* E. Sartorelli, and M. Tomasini. *Med. lavoro*, February, 1959, 50: 114–119. (*U. of Milan, Italy)

The data presented refer to a 54-year-old patient with reticular micronodular silicosis who underwent pneumectomy for an abscess of the left upper lobe; all of the measures were taken 4 years after the operation. Cardiac catheterization revealed hypertension of the pulmonary circulation, associated with a marked increase of the arteriolar pulmonary resistance, neither related to arterial hypoxemia nor changeable by the uptake of pure oxygen. According to these results, the pulmonary hypertension should have an anatomic origin, namely, for the reduction of the capillary bed and a diminished elasticity of the arteriolar wall; both processes are present in silicosis.

I. Archetti

Pulmonary Resection for Tuberculosis. Medical and Social Results in 122 Students Followed at the "Post-cure Universitaire" of Grenoble from 1950–1957 (in French). D. Neel* and C. Tran Hiep. *Rev. tuberc., Paris*, January, 1959, 23: 123–133. (*Grenoble, France)

Pulmonary resection was performed in 122 students between the ages of 18 and 35 from 1950 to 1957. There were 10 pneumonectomies, 64 lobectomies, and 52 segmental resections. Surgical indications for far advanced cases with extensive cavitation decreased from 20 per cent of all cases before 1954 to 8 per cent thereafter. Antituberculosis chemotherapy for more than 5 months had been given to 25 per cent of the patients in 1953, 56 per cent in 1954, and 75 per cent in 1956.

The majority of postoperative complications occurred early (44 cases); 7 complications occurred more than 6 months after operation. The most important of the early complications were: bronchopleural fistulas (10), alveolar air leak with hydropneumothorax (10), postoperative hemorrhage (6), large pleural effusions (7), empyema (3), and pulmonary embolism (1). The duration of chemotherapy prior to resection did not seem to play a definite role in the prevention of bronchopleural fistulas. Postoperative complications did not show any diminution in recent years in spite of progress in medical treatment and surgical technique.

There were 17 relapses (13.9 per cent of all patients with resection), relapse being defined as active tuberculosis developing 3 months after resection; 4 of the relapses occurred from 3 to 6 months after operation, 13 at a later date (the latest, 33 months after resection). The incidence of relapse was in inverse proportion to the duration of postoperative chemotherapy. The prognosis of relapses was, on the whole, favorable, with the exception of 1 death due to extrapleural pneumothorax. In order to minimize pleural and functional sequelae, patients were started on functional re-education and physiotherapy very early after surgery. As of the spring of 1958, 73 per cent of the patients had practically normal chest roentgenograms; 27 per cent had stable nodular and strand-like densities. One-third of the patients had pleural sequelae. Of the 122 patients, 117 had resumed normal full-time activity.

V. Leites

Surgical Exeresis in the Treatment of Pulmonary Tuberculosis (in Italian). G. D. Bianchera,* G. Bozzetti, and G. Ferrara. *Ann. med. Sondalo*, January-February, 1959, 7: 18–32. (*V. Sanatoriale, Sondalo, Italy)

The data presented are based upon 304 operations for pulmonary tuberculosis. The patients were divided into: favorable (cases with unilateral and circumscribed lesions less than 2 cm. in diameter), moderately severe (cases with unilateral lesions more than 2 cm. in diameter, involving 1 or more lobes), and very severe cases (those with lung destruction accompanied by empyema, bilateral lesions, et cetera).

The results, expressed in percentage, were as follows: good in 100 of the first group, 75.3 of the second, and 59.4 in the third; incomplete in 0 of the first, 15.3 of the second, and 23.6 of the third group; mortality was 0 in the first, 9.3 in the second, and 16.9 in the third. In 161 cases (63 per cent), different procedures for obliterating the residual cavity were used.

I. Archetti

Fibroma of the Trachea. M. A. Miller* and G. A. Toma. *Brit. J. Dis. Chest*, April, 1959, 53: 177–180. (*Whittington Hosp., London, England)

A case is presented of fibroma of the trachea with progressive dyspnea and widespread surgical emphysema with a marked expiratory wheeze; but there were no added sounds in the chest. The wheeze heard at the open mouth, not at the chest wall, is usually the earliest symptom, and this wheeze, described as an "asthmatoid wheeze," has often led to an erroneous diagnosis of asthma. Immediate diagnostic bronchoscopy in such cases is the only means of determining the character of the lesion or substance causing the obstruction. The literature is reviewed of the tracheal tumors reported up to December, 1945.

M. J. Small

Primary Fibrosarcoma of the Lung in a Young Child: A Case Treated by Lobectomy and Cobalt Therapy. F. S. GERBASI,* A. M. MARGILETH, and R. S. KIBLER. *J. Pediat.*, April, 1959, 54: 488–495. (*Alexander Blain Hosp., Detroit, Mich.)

Primary fibrosarcoma of the lung in a 33-month-old child is reported. This is the youngest case of this type of sarcoma of the lung ever recorded, and the only case recorded in a child treated by lobectomy and cobalt therapy. This was a very anaplastic fibrosarcoma which responded poorly to the above-mentioned therapy. Excision of the lesion relieved the boy's dyspnea temporarily, and the cobalt therapy had little or no therapeutic value. The patient developed extensive metastases and died 13 months after the onset of symptoms (Authors' summary).

M. J. SMALL

Pulmonary Resection for Mineral Oil Granulomas. L. D. GUIDRY,* O. T. CLAGETT, J. R. MCDONALD, and O. W. KINCAID. *Ann. Surg.*, July, 1959, 150: 67–76. (*Mayo Clinic, Rochester, Minn.)

Eleven cases in which pulmonary resection had been performed for mineral oil granulomas of the lung at the Mayo Clinic since 1947 were studied with special reference to the problems of diagnosis and treatment. Exploratory thoracotomy is advocated for disease indistinguishable from bronchogenic carcinoma regardless of whether mineral oil droplets can be detected in the sputum. Excision is favored for symptomatic or extensive pulmonary involvement when this can be accomplished without excessive sacrifice of normal lung tissue. The establishment of diagnosis should precede any contemplated resective procedure (Authors' summary).

M. J. SMALL

Study of Surgically-excised Pulmonary Granulomas. E. L. SEGAL,* G. F. STARR, and L. A. WEED. *J. A. M. A.*, May 30, 1959, 170: 515–522. (*U. of Minnesota, Rochester)

By the application of Gomori's methenamine-silver nitrate stain (GMS) to 138 surgically-excised pulmonary granulomas, it was possible to identify "structures" resembling *Histoplasma capsulatum* or *Coccidioides immitis* in 55.9 per cent. Despite extensive efforts, it was not possible to obtain fungal growths from these structures, which are thought to be nonviable fungi that have lost their ordinary stainability. With the GMS stain, fungi can be found in tissue sections more rapidly and under lower magnification than by other staining methods.

H. ABELES

PUBLIC HEALTH AND EPIDEMIOLOGY

The Statistical Investigation of Smoking and Cancer of the Lung. J. BERKSON.* *Proc. Staff Meet. Mayo Clin.*, April 15, 1959, 34: 206–225. (*Mayo Clinic, Rochester, Minn.)

The author, a statistician, in an article which is difficult to abstract, questions the validity of recent statistics which link, in a causative fashion, cigarette smoking and the development of bronchogenic carcinoma. It is pointed out that, although the death rate for cancer of the lung is higher among smokers than nonsmokers, the statistical conclusions have not been fully corroborated by experimental and direct observational studies. He also states that the sampled populations studied in two recent statistical surveys are not representative of the reference population from which these samples were drawn, in that the general and specific death rates are low compared with corresponding death rates for the general population. Although there is a graded increase of death rate from cancer of the lung with increase in amount of smoking, there is also an increase of death rates for many other nonrelated diseases. In one study, only 13.5 per cent of these excess deaths were due to lung cancer, while 86.5 per cent were due to other causes, particularly coronary artery disease.

The author believes that the most important finding of these statistical studies is not that there is an association between smoking and cancer of the lung, but that there is an association between smoking and deaths from all causes generally. However, he questions the validity of this statistical association on the grounds that: (*1*) the datum of smoking history is subject to wide variation, (*2*) the cause of death as recorded on death statistics may be unreliable, and (*3*) in none of the investigations does the sample studied constitute a really adequately large proportion of the reference population. He also suggests that persons who are mild- or nonsmokers may be constitutionally disposed to longevity and low death rates, or that smoking in some manner may speed up the normal process of aging with the resulting general increase in death rates.

E. A. RILEY

Erythema Component of the Primary Mantoux Response. F. O'GRADY.* *Brit. J. Dis. Chest*, April, 1959, 53: 181–186. (*Middlesex Hosp., London, England)

Various forms of erythema response to tuberculin are described and the reasons for believing this response to be partly independent of induration

are given. There was no evidence that a particular form of erythema response occurred in the presence of active tuberculosis, but it appeared that florid erythema was seen more commonly in patients with active disease. Erythema was rare in those who failed to develop induration to 100 tuberculin units, and it is suggested that the nonspecificity of the erythema response to tuberculin has been overemphasized. In the present series, in 136 negative reactors, lone erythema more than 10 mm. in diameter was seen in the proved absence of active tuberculosis in only 2 cases. The view that erythema is an entirely nonspecific response of haphazard appearances seems very difficult to justify.

M. J. Small

Changes in Erythema as the Index of Sensitisation and Activation. F. O'Grady.* *Brit. J. Dis. Chest*, April, 1959, 53: 187–192. (*Middlesex Hosp., London, England)

The frequency with which increases in erythema and induration gave evidence of tuberculin sensitization in the presence and absence of active tuberculosis has been compared. Failure to develop increased induration on retesting—in itself an unreliable guide—was a better index of the presence of active tuberculosis than any test based on changes in erythema. The frequency with which increased and decreased erythema were encountered suggests that these changes were the result of chance. When, as a result of retesting, induration reappeared at the site of a previous test, it was rarely accompanied by erythema. The stimulus for the development of induration and erythema appears, therefore, to be the same, but the difference in the ability of tuberculin and tuberculous infection to reinstate erythema at an old tuberculin site again emphasizes that their roles in invoking sensitization are not identical.

M. J. Small

An Epidemic of Pleurisy with Effusion in Bitlis, Turkey. Study of 559 Case Reports (in French). F. Alptekin. *Presse méd.*, April 25, 1959, 21: 851–852.

An epidemic of pleurisy with effusion was observed in Bitlis, Turkey, in 1955. All cases showed an acute clinical course with fever. All of the 559 cases reported here presented a picture different from any known pleurisies or respiratory diseases. In all cases, a pleural effusion appeared at the base. The fluid showed a high count of monocytes exceeding all other cellular elements. There was a long convalescence of up to 3 or 4 months. Most cases occurred at the end of winter and in spring. The incidence was highest in overcrowded barracks. It seems very probable that this disease, which is called "epidemic pleurisy," is viral in origin.

E. Lyon

Two Cases of Silicosis Associated with Scleroderma (in Italian). F. Bellini* and E. Ghislandi. *Med. lavoro*, January, 1959, 50: 63–70. (*U. of Milan, Italy)

Two cases of silicosis associated with scleroderma, in patients 33 and 55 years old, are briefly described. Tuberculosis was excluded as a diagnostic possibility, and in both cases the worsening of silicosis seemed to be caused by the sclerodermic process.

The roentgenographic pattern of the 2 cases are given in detail: in the first case the roentgenogram was characterized by an intense reticulation with diffuse micronodulation, parenchymal shadow of the right basis of the lung, and egg-shell calcification of the lymph nodes; in the second patient there were subpleural condensation in both apical regions, areas of emphysema in both mean pulmonary fields, and rough pleural stripes in the mean basal regions.

I. Archetti

Epidemiologic Importance of Isoniazid-resistant Tubercle Bacilli (in Turkish). N. Karasu,* O. Oger, A. Gursel, and A. H. Gurses. *Tuberk. Toraks*, January–April, 1959, 7: 55–61. (*U. of Ankara, Turkey)

All of the tubercle bacilli recovered from 27 untreated cases of pulmonary tuberculosis were found to be isoniazid-susceptible. Nineteen of 100 previously treated cases were found to have isoniazid-resistant tubercle bacilli in their sputum. No new case of tuberculosis was discovered among the contacts of isoniazid-resistant cases. Thirty-five per cent of the contacts of patients with isoniazid-susceptible bacilli showed evidence of recent tuberculous infection.

S. Solu

Forcible Detention of Patients with Active Tuberculosis. R. Glass.* *Pub. Health Rep.*, May, 1959, 74: 399–404. (*N. Y. C. Health Department, N. Y.)

In June, 1955, the New York City Department of Health in cooperation with the Department of Correction set aside beds at the Rikers Island Hospital for enforced hospitalization of recalcitrant male patients with active pulmonary tuberculosis who were a danger to public health. A total of 46 patients, including one who was readmitted, were put on the detention service from its initiation in July, 1955, through December 31, 1957. Of these, 20 became arrested cases of pulmonary tuberculosis, either while confined to the

detention service or after transfer to a non-detention hospital. By December 31, 1957, 18 patients were still hospitalized on the detention service or in non-detention hospitals. Seven of these men showed improvement, bacteriologically and by chest film, quantitatively and qualitatively, which may permit expectation of control of their condition in the future under continued hospital treatment. The 46 recalcitrant patients treated in about 18 months represent a small proportion of the number of persons in the area with active pulmonary tuberculosis requiring hospitalization. Stimulated by results during this limited period, preparations are being made for expansion of the forcible detention program.

E. DUNNER

Respiratory Virus Infection in the Glasgow Area. N. R. GRIST.* *Brit. M. J.*, April 4, 1959, No. 5126: 900–901. (*Ruchill Hosp., Glasgow, Scotland)

More than 1,000 cases of acute respiratory illness were investigated virologically during two years. Ten cases of adenovirus infection were detected; one of these presented the features of glandular fever. Ten cases developed complement-fixing antibodies to Sendai virus; of 4 such cases tested with mumps antigens, only 2 developed antibodies—1 to both "V" and "S" antigens, and 1 to "V" antigen alone. These findings suggest infection with myxovirus related but not necessarily identical with Sendai virus. Type A influenza viral infections were detected during the autumn epidemic of 1957 and in the early months of 1958. No antigenic variation of the virus was detected. Three cases of encephalitis with serologic evidence of influenza were encountered during the outbreak (after author's summary).

E. A. RILEY

ROENTGENOGRAPHY

Pulmonary Arteriovenous Aneurysm. P. ELLMAN* and A. HANSON. *Brit. J. Dis. Chest*, April, 1959, 53: 165–172. (*Plaistow Hosp., London, England)

The literature relating to pulmonary arteriovenous aneurysms is surveyed. A case is reported of pulmonary arteriovenous aneurysm complicated by active pulmonary tuberculosis, chronic bronchitis, and emphysema, successfully treated by resection of the aneurysm.

The late onset of signs and symptoms in this case is discussed in relation to the coexistent pulmonary pathology. Clubbing was first observed at the age of 40, a transient cerebral episode at 45, epistaxis at 51, and cyanosis was not apparent until the age of 52. Initially the degree of clubbing may well have been accepted as commensurate with the patient's chronic bronchitis and associated tuberculosis. It was only after the development of bronchospasm that a slight increase in the size of the lung tumor could be detected, and it was presumably during this period that the shunt became sufficiently large to produce the typical cyanotic and polycythemic changes.

The importance of tomography and angiography as preoperative diagnostic procedures is considered. Although tomography can often give confirmative evidence without angiography, the latter is preferable in that it gives information not only about the roentgenographically obvious lesions, but also about small lesions which might easily be overlooked on a straight film or in localized tomography. Multiple aneurysms have been reported in over 30 per cent of the recorded cases and, in order to gain a really satisfactory result from surgery, it is essential that all clinically significant lesions be eradicated, even if this involves more than one operation; otherwise some cyanosis may remain, or cyanosis may again develop after the patient has apparently been cured.

That surgery is advisable is indicated by the relatively high incidence of hemoptyses, hematemeses, and epistaxes which may prove sufficiently severe to be fatal. Cerebral thrombosis due to increased blood viscosity is also a hazard, and neurologic disturbances are relatively common. Dyspnea is often dramatically relieved by surgery, and there is no doubt that the return to normal color is, for the patient, a satisfactory psychologic as well as physiologic outcome. The hazards and complications occurring in the surgical approach to pulmonary arteriovenous aneurysms are reported.

M. J. SMALL

Communication Between the Anterior Aortic Sinus and the Right Ventricle Diagnosed by Thoracic Aortography. J. PHILIPSON* and G. F. SALTZMAN. *Acta radiol.*, April, 1959, 51: 283–288. (*Sodersjukhuset, Stockholm, Sweden)

The value of thoracic aortography in rare heart and great vessel malformations with left to right shunt near the aortic valves is pointed out. Mention is made that, by thoracic aortography, the best anatomic details are achieved, and a diagnosis is possible when a distinction is to be made between an aortic septal defect, a ventricular septal defect with aortic insufficiency, and a communication between the aortic bulb and the right ventricle. Clinical data on a case report is given.

E. SHABART

An Evaluation of the Pharmacological Hazards Resulting from Use of "Visciodol" in Bronchography. P. M. JOHNSON* and G. I. IRWIN. *Radiology*, June, 1959, 72: 816–828. (*U. of N. Carolina, Chapel Hill)

Thirty-five patients were studied for sulfanilamide absorption and methemoglobin formation after bronchography with Visciodol® (an iodized oil–sulfanilamide suspension, each cubic centimeter of which contains 0.32 gm. of sulfanilamide employed solely as a viscosizing agent). Using accepted doses of the contrast medium, bronchography with Visciodol invariably produced sulfanilamidemia, the magnitude of which was dependent upon the volume given, the degree of alveolar spillage, and the amount ingested. This sulfanilamidemia exceeded the therapeutic blood sulfanilamide level and persisted for at least 24, and usually 48, hours. Methemoglobin formation occurred in 29 patients and produced overt cyanosis at higher levels. It is concluded that the use of Visciodol carries a small but unjustifiable risk.

For physicians who may continue to use this medium, it is recommended that Visciodol should not be employed when the patient is unable to cooperate, has unsuppressed cough, or gives a history of sulfanilamide reaction. Dosage should be kept as low as possible. In patients with respiratory insufficiency, consideration should be given to prophylactic pretreatment with methylene blue. Cyanosis after Visciodol bronchography should be considered due to methemoglobinemia unless there are compelling signs of another cause. Intravenous administration of methylene blue is both a therapeutic test and the treatment of choice.

W. J. STEININGER

Image Contrast. P. VUORINEN.* *Acta radiol.*, April, 1959. 51: 305–307. (*Wenner-Gren Research Lab., Norrtulls, Stockholm, Sweden)

Metal phantoms are commonly used as test objects in tomographic experiments, especially when layer thickness is to be measured. It has been shown that a metal object may be moved several millimeters during exposure; but the resultant motional blurring is evident only if the contrast is not unduly changed. In order to test this principle of tomography, an experiment is carried out and discussed. The diagrams are presented. The experiment appears to point out that due consideration to the image contrast must be observed or the result may otherwise be misleading.

E. SHABART

Mass Radiography and Cancer of the Lung. E. POSNER,* L. A. MCDOWELL, and K. W. CROSS. *Brit. M. J.*, May 9, 1959, No. 5131: 1213–1218. (*Stoke on Trent Mass Radiography Unit, Staffordshire, England)

A series of 238 cases of cancer of the lung found by mass roentgenography is compared with cases referred to units by general practitioners. The over-all 2-year survival rates were 20 per cent of cases referred by general practitioners and 25 per cent of cases picked up on routine survey. Among the resected cases, the 2-year survival rates were 44 per cent and 38 per cent, respectively. Forty-four per cent of the routine survey group were found resectable, compared with 30 per cent of the general practitioner group. Squamous neoplasms proved to be more resectable than the undifferentiated tumors. More than $\frac{1}{3}$ of patients seen by general practitioners were referred for chest films within the first month of consultation with their doctors, and at least $\frac{2}{3}$ within the first 2 months. Sixty-seven per cent of the cases were operated upon within 2 months of roentgen examination, but the figures failed to show a direct relationship between early referral and increased resectability. Only 15 per cent of the routine survey group were asymptomatic.

E. A. RILEY

Method for Irradiation of Parasternal Lymph-Node Metastasis. F. EDSMYR* and R. WALSTAM. *Acta radiol.*, April, 1959, 51: 308–320. (* Karolinska Sjukhuset, Stockholm, Sweden)

This paper discusses the recent attention to carcinoma of the mammary glands and the risk of metastasis within the parasternal region. A survey of the literature reveals the possibility of parasternal metastasis being greater if the primary tumor is situated in the medial or central part of the breast. Most metastases have been found in the second to fourth intercostal spaces.

A detailed discussion is given on the use of conventional roentgen radiation and teleradium therapy. Two groups of patients receiving Co^{60} radiation were studied. The first comprised cases treated with ordinary treatment tubes. The second group consisted of those cases treated with a specially designed parasternal tube. The overlapping field technique was employed. A series of treatments is usually given within 12–20 days.

The time of observation is believed to be too short to allow adequate estimation of the end results of the treatments. A temporary leukopenia was observed in a few cases. Skin reaction was considered unimportant or moderate. A few cases showed serous dermatitis which was most preva-

lent in the axilla and fossa supraclavicularis. Only 19 per cent of the patients had any complaints during the course of the treatments.

E. SHABART

CHEMOTHERAPY

4-Aminosalicylic Acid Blood Levels Following Oral Administration of Sodium 4-Aminosalicylate and Calcium 4-Benzamidosalicylate. D. J. DRAIN,* R. LAZARE, and K. TATTERSALL. *Tubercle*, June, 1959, 40: 201–204. (*Smith and Nephew Research Ltd., Hunsdon Lab., Ware, Hertfordshire, England)

Previous comparisons of PAS blood levels produced by sodium PAS and calcium B-PAS have been based on the use of chemical estimations. It has long been realized that chemical methods available for the estimation of PAS are not specific and measure any aromatic amine. High blood amine levels obtained after sodium PAS treatment are not solely PAS, but include much biologically inactive material. It would seem therefore that, in measuring PAS blood levels, microbiologic methods are to be preferred in that their greater specificity outweighs the disadvantages imposed by the limitations of accuracy inherent in such procedures.

The results presented here, although based on very small numbers of cases, indicate that, if such a basis of comparison is used, the PAS blood levels after a period of treatment with calcium B-PAS, although lower than those produced by sodium PAS, do not differ by so great a margin as has been previously supposed. Further work in progress is designed to answer some of the questions posed by these preliminary observations and may resolve doubts concerning the clinical efficacy of calcium B-PAS, which have been based on considerations of PAS blood levels.

M. J. SMALL

Treatment of Chronic Pulmonary Heart Disease with Thioctic Acid (in Italian). L. CHIANCA* and E. CICOIRA. *Minerva med.*, September, 1958, 49: 3466–3471. (*Osp. Riuniti, Naples, Italy)

Significant results were obtained in 40 patients, 20 with pulmonary heart disease associated with either moderate or severe cardiac failure, who were daily given 20 mg. of thioctic acid, the co-enzyme whose main property is to favor the synthesis and transfer of the acetylic radicals.

Therefore, it was suggested that this chemical has an elective therapeutic action in conditions of congestive failure, which occurs in pneumocirculatory syndromes. This therapeutic action should be due to the action of the substance of favoring the utilization of the oxygen both at the cellular level and in the lung, of regulating the salt and water exchange and, finally, to its protective effects on the liver and the kidney.

I. ARCHETTI

First Clinical Observations on the Treatment of Pulmonary Tuberculosis with Alpha-Ethyl-Thioisonicotinamide (in Italian). G. DADDI,* M. CORDA, C. GRASSI, and C. SACERDOTI. *Gior. ital. tuberc.*, November–December, 1958, 12: 305–311. (*U. of Milan, Italy)

A total of 63 patients with different forms of pulmonary tuberculosis were given 1.5 gm. of α-ethyl-thioisonicotinamide (ethylisotiamide) daily during the first 5 days of treatment, then 1 gm. for a period of about one to two months. Roentgenographic and bacteriologic controls were periodically taken.

The therapeutic effects of the drug were often good, so that at the end of treatment most of the patients showed clear improvement. There was frequently a definite decrease in the number of tubercle bacilli in the sputum; bacilli resistant to other antimicrobials very often disappeared and, in about ⅓ of the cases, resistance to this new drug became manifest. Symptoms of intolerance were observed in 45 per cent of the patients: they were mild in men and more serious, but not severe, in women.

I. ARCHETTI

Preliminary Study of the Effect of Alpha-Ethyl-Thioisonicotinamide (1314 Th) in Chronic Pulmonary Tuberculosis (in French). G. BROUET,* J. MARCHÉ, N. RIST, J. CHEVALLIER, and G. LE MEUR. *Rev. tuberc, Paris*, January, 1959, 23: 36–66. (*Paris, France)

1314 Th was synthesized by Libermann, N. Rist, and F. Grumbach. It is not a simple derivative of isoniazid because it is effective against isoniazid-resistant strains, and bacilli resistant to 1314 Th remain susceptible to isoniazid *in vitro* and *in vivo*. It is effective against strains resistant to all other antituberculosis drugs currently in use, except the thiosemicarbazones. The clinical trials of 1314 were started in the spring of 1957. The present study comprises 78 patients, 43 of whom had recent pulmonary tuberculosis with known duration of less than 6 months; in 55 the disease was of long standing.

The drug was often poorly tolerated, producing gastrointestinal troubles (anorexia, nausea, gastralgia) which often were the cause of inability to gain weight, or even of weight loss (24 patients). Of 79 patients, 6 were able to take the drug regu-

larly without major inconvenience; in 29, tolerance was poor; in 7, very poor; and in 5, the drug had to be discontinued. Renal and hepatic function showed no disturbance under treatment. No neurotoxicity was observed when 1314 was given alone, but when added to isoniazid or cycloserine in alcoholics or patients with previous neuropsychiatric disturbance, convulsions and polyneuritis were observed (4 patients).

Of 23 patients with recent minimal and moderately advanced tuberculosis treated with 1314 alone, 10 (43.5 per cent) showed marked and very marked roentgenographic clearing and reversal of infectiousness; 2 patients failed to improve; and moderate roentgenographic improvement and reversal of infectiousness were seen in 7. Of 32 patients with active tuberculosis of long duration in whom 1314 was added to various other antituberculosis agents, there were 6 cases of reversal of infectiousness and marked or very marked roentgenographic clearing; 12 other patients showed sputum conversion with partial roentgenographic clearing. However, the effect of the drug as observed during the period of this study was not constant. It should not be used alone in the "treatment of attack" of recent pulmonary tuberculosis.

V. LEITES

Short-term Study of Treatment with Alpha-Ethyl-Thioisonicotinamide (1314 Th). A Preliminary Report (in French). THE FOUNDATION OF STUDENT SANATORIUMS AND THE DISPENSARY GUY RENARD, PARIS. *Rev. tuberc., Paris*, February-March, 1959, 23: 185–203.

A preliminary report is presented on 62 patients with pulmonary tuberculosis, mostly of long duration, who were treated with 1314 Th (α-ethyl-thioisonicotinamide). Adequate bacteriologic and roentgenographic data are available for only 32 patients followed for more than 3 months. Of the 21 patients who had infectious sputum prior to therapy, 10 showed reversal of infectiousness by smear and culture. Seven of these patients with sputum conversion had extensive, severe tuberculosis. Of the 32 patients, 21 showed roentgenographic improvement which was marked in 10; 10 patients remained stationary; 1 showed worsening.

Toxic manifestations developed in 55 of 62 patients: 42 patients had gastric disturbances consisting most often of epigastric pain and burning. These symptoms occurred intermittently at irregular intervals; they varied in intensity but were mostly mild, permitting continuation of treatment. In 3 cases treatment had to be discontinued, in 9 the dose had to be lowered or the drug stopped temporarily. Psychologic disturbances occurred in 29 of 62 patients and were mainly of the depressive type. Sixteen patients had acneiform skin eruption during treatment. Menstrual anomalies attributed to the drug were noted in 12 of 23 female patients. All toxic reactions due to 1314 disappeared rapidly after cessation of treatment. In the entire series there were no permanent sequalae of toxicity.

V. LEITES

Current Status of Amphotericin B in the Treatment of the Systemic Fungus Infections. V. D. NEWCOMER,* T. H. STERNBERG, E. T. WRIGHT, and R. M. REISNER. *J. Chronic Dis.*, April, 1959, 9: 353–374. (*U. of California Med. Center, Berkeley)

Amphotericin B is a new polyene antifungal antimicrobial which, *in vitro*, inhibits a wide variety of pathogenic and nonpathogenic fungi including *Blastomyces dermatitidis*, *Candida albicans*, *Cryptococcus neoformans*, *Histoplasma capsulatum*, and *Coccidioides immitis*. The intravenous route of administration appears to be the most effective. Twenty-two patients with deep mycoses were treated with intravenous amphotericin B in dosages of 0.7 to 1.4 mg. per kg. daily or on alternate days over a period of 2 weeks to 4 months. It appeared that this drug was of significant value in the treatment of systemic mycoses.

C. A. ROSS

Chelating Agents in the Therapy of Beryllium Poisoning. R. CASH,* R. I. SHAPIRO, S. H. LEVY, and S. M. HOPKINS. *New England J. Med.*, April 2, 1959, 260: 683–866. (*Children's Hosp. of Philadelphia, Pa.)

Trisodium monohydrogen EDTA (ethylene diamine tetra-acetic acid), a polyamino carboxylic chelating agent, was used in an attempt to increase the renal excretion of beryllium in 2 cases of chronic beryllium pneumonoconiosis. It was found that, within the limits of the study, this agent enhanced the renal excretion of beryllium. Calcium EDTA was found to have a similar effect. Since this was a short-term study, designed only to test the effectiveness of this chelating agent in mobilizing beryllium deposited in the body tissues, the effect of this treatment on roentgen changes or pulmonary function could not be evaluated. These aspects are to be reported on at a later date.

Emphysema and diffuse fibrosis are presumably much more important than is granuloma from the standpoint of symptomatology, but it is suggested that long-term intermittent therapy with a chelating agent such as EDTA may so deplete the body stores of beryllium ion that the clinical course or progress of the disease may be favorably affected.

M. J. SMALL

Ambulant Chemotherapy: A Practical Advance in the Treatment of Pulmonary Tuberculosis. S. E. Slade.* *Brit. J. Dis. Chest*, April, 1959, 53: 193–196. (*Dept. of Pub. Health, Govt. of Western Australia)

The nature and advantages of ambulant chemotherapy are described, and 40 consecutive personal cases are analyzed. The suitability of this type of treatment is suggested when roentgenographic evidence of disease is associated with the absence of tubercle bacilli in the sputum of an intelligent patient without signs of toxemia, i.e., no constitutional disturbance and a normal sedimentation rate. Twelve patients in this series fell into this category; in addition 28 had tubercle bacilli demonstrated by laryngeal swab or sputum examination, but refused institutional treatment although it was indicated and advised. Twenty-five were converted to the sputum-negative state. One patient died during treatment. One patient was uncontrolled and had atypical organisms. It is submitted that ambulant chemotherapy in suitable cases has domestic, social, and financial advantages over conservative methods.

M. J. Small

Short-Term ACTH Therapy for Desensitisation Against Anti-Tuberculosis Drugs. S. K. Gupta* and H. D. Palmer. *Brit. J. Dis. Chest*, April, 1959, 53: 197–201. (*Harts Hosp., Woodford Green, Essex, England)

The occurrence of allergic reactions to antituberculosis drugs is well known. The incidence varies in different centers, but these hazards are particularly common during the first 3 months of treatment. Fortunately, desensitization is possible in most cases, but the current methods are unsatisfactory. Hence, a short scheme using a high dosage of corticotropin (ACTH) was utilized.

The drugs were withdrawn in each case as soon as an allergic state was diagnosed, because an inadvertent use of the offending drug at this time might cause serious results. As the reactions settled, either an intramuscular, intracutaneous, or oral test dose was administered to determine the offending drug. The importance of this procedure is evident in one patient who was previously "desensitized" against streptomycin, although he was susceptible to *p*-aminosalicylic acid. Sixty units of corticotropin daily were sufficient in most cases. On the sixth day it was usually possible to discontinue the corticotropin cover. In every case, however, the usual 3 drugs were given concurrently.

None of 12 cases treated had any serious complication. Following withdrawal of corticotropin, 2 elderly men developed muscular aches, 1 sufficiently severely to require analgesics. Many patients rapidly gained weight which was subsequently lost by diuresis. Among the treated cases, 1 had previous history of glycosuria, 2 had hypertension, while another had a history of peptic ulceration. Though unexplained, the initial superiority of corticotropin over cortisone and prednisone has been noticed by several observers. Isolated records of desensitization with prednisone have been reported, but the results are not promising.

Short-term corticotropin therapy has many advantages. The course is brief so that no special precaution is necessary. Complications are few and mild. Desensitization to 2 or more drugs may be carried out simultaneously. The patient has no break in the treatment as the drugs are administered concurrently. No drugs need to be withdrawn, so the morale of the patients remains high. The scheme is adjustable. Higher dosage of corticotropin may be instituted at any time, or the course may also be prolonged. No adverse effect on the disease was noted in any case. Cases showing signs of anaphylaxis probably need more care, and the full dosage may be restored on the fifth day or later.

M. J. Small

Comparison of Results Obtained with Different Doses of Isoniazid (5 and 15 Mg. per Kg. of Body Weight). A Study of 204 Cases of Pulmonary Tuberculosis in the Adult (in French). É. Bernard,* A. P. Jarniou, L. Israel, M. Enjalbert, and Mlle Ch. Fabre. *Rev. tuberc., Paris*, January, 1959, 23: 22–34. (*Paris, France)

This study includes 204 patients with pulmonary tuberculosis treated from June, 1957 through September, 1958. Excluded from the study were patients with previous chemotherapy, tuberculous meningitis requiring combined therapy, and alcoholic neuritis. The patients were assigned high and low doses of isoniazid by random allocation: 15 mg. per kg. of isoniazid in 3 divided doses, and 15 gm. of PAS intravenously, as well as 200 mg. of pyridoxine 3 times weekly, or 5 mg. per kg. of isoniazid and 15 gm. of PAS intravenously.

Each patient was observed for 3 months. Marked and very marked roentgenographic improvement was observed in 50.8 per cent of the patients given high doses, and in 38.8 per cent of the patients given lower doses. There were 12.2 per cent deteriorations among the patients given high doses, and 17.7 per cent among the controls. Cavitary lesions showed marked and very marked improvement in 60.2 per cent given high doses, and 40.3

per cent of those given low doses. After 3 months of treatment, 18 per cent of the patients still had infectious sputum in the high-dosage group, as compared to 26 per cent in the control group. The high doses were well tolerated; treatment had to be interrupted in only 1 case because of an excitatory effect which subsided after withdrawal of the drug.

V. LEITES

Psychic Disturbances Provoked by Isoniazid. Their Correlation with the History of Psychic Disturbances and the Level of Free Isoniazid in the Blood of Patients (in French). A. LEPEUPLE,* R. THIBIER, J. N. VIVIEN, J. GROSSET, and G. CANETTI. *Rev. tuberc., Paris,* February-March, 1959, 23: 214–230. (*Paris, France)

Among 621 patients (students treated in the French Student Sanatoria), 7 per cent gave histories of mental disturbance; in 4.6 per cent this was severe (psychosis or severe neurosis). The total incidence of psychologic disturbance due to isoniazid was 3.8 per cent (24 patients); none of these patients gave a history of psychosis. However, of 44 patients with histories of neurosis of varying intensity, 15 (1 of 3) showed psychic disturbances attributed to isoniazid. Among 577 patients with negative past histories, isoniazid caused psychic side effects in only 9 (1 of 64). In 16 of the 24 cases, psychologic side effects due to the drug occurred within the first 6 months of treatment, and in ¼ of the cases, after one year of treatment.

Most patients received isoniazid in a dosage of 5–6 mg. per kg. There did not seem to be any correlation between daily dosage and side effects. The mean blood levels of free isoniazid were 1.47γ in patients without side effects, and 1.65γ in those with psychologic disturbance. The nature of the side reactions is described as follows: (*1*) mild reactions (unexplained euphoria and exaltation, transient memory defects, especially for recent events, diminution of self control) 9 cases; (*2*) moderately severe reactions, mostly of the manic or manic depressive type, 11 cases; (*3*) severe reactions, characterized by confusion, motor agitation, and subsequent amnesia, 4 cases. The patients were not amenable to psychotherapy because of emotional instability. However, the disturbances disappeared rapidly after discontinuance of isoniazid.

V. LEITES

Bactericidal Action of Kanamycin on *Mycobacterium Tuberculosis in Vitro* (in Italian). V. NITTI.* *Arch. tisiol.*, January, 1959, 14: 37–49. (*U. of Naples, Italy)

The bactericidal action of kanamycin on *M. tuberculosis* strain H37Rv, grown in Dubos medium with Tween® 80, was tested using 2-5-10-50γ of kanamycin per ml.; samples were taken after 1, 2, 4, and 10 days of contact, and counts of surviving bacteria were done in Löwenstein-Jensen solid medium in Petri dishes after 4 weeks of incubation at 37°C.

Kanamycin had a clear effect even at the lowest concentration, and reached its optimum at 10γ per ml.; the action was evident from the first day and became progressively more manifest in the cultures with the highest concentrations, while bacteria started to grow again in the cultures containing 2γ per ml. Normal bacterial cells could be found together with resistant mutants in each experiment; they survived the action of the antimicrobial, which appeared to be purely bacteriostatic.

I. ARCHETTI

Therapeutic Variations of PAS Levels in the Blood and Their Influence on the Metabolism of Isoniazid (in French). G. FAVEZ,* G. BORSARI, and H. LAUENER. *Rev. tuberc., Paris,* January, 1959, 23: 136–141. (*Lausanne, Switzerland)

During treatment with isoniazid and PAS, the inactivation of isoniazid depends, at least partly, upon the transformation of the para-aminosalicylates. The minimal dose of calcium benzoyl-PAS still capable of increasing the level of free isoniazid in relation to total isoniazid is close to 4 gm. A dose of 2 gm. of calcium benzoyl-PAS administered with each of 3 daily doses of 100 mg. of isoniazid proved unable to significantly reduce acetylation of isoniazid. An equimolecular dose of PAS (525 mg.) and isoniazid (475 mg.) was equally ineffective. The infusion of 20 gm. of sodium PAS decreases the inactivation of isoniazid during the first hours more than does a single dose of 6.7 gm. Long lasting infusion (6 hours) is less effective in protecting isoniazid from acetylation than is short lasting infusion.

V. LEITES

PAS Toxicity (in Turkish). T. FIRATLI,* Y. AKSU, T. INANC, and I. ULGENALP. *Tuberkuloz*, January, 1959, 12: 42–55. (*Ataturk Sanatorium, Ankara, Turkey)

A case of death due to PAS toxicity is presented. The cause of death, as determined at autopsy, was acute hemolytic anemia and parenchymal organ necrosis following a toxi-allergic reaction in a PAS-sensitized subject.

S. SOLU

Streptomycin Poisoning in Renal Failure. K. D. G. EDWARDS* and H. M. WHYTE. *Brit. M. J.*, March 21, 1959, No. 5124: 752–755. (*Sydney Hosp., Australia)

Five instances are described of streptomycin poisoning in patients with renal insufficiency. In

2 patients with acute oliguric renal failure, the symptoms of vestibular damage appeared 24 days after the last dose of streptomycin and after diuresis had occurred. *In vitro* tests showed that streptomycin can be removed from blood by dialysis, and early treatment with an artificial kidney is recommended for oliguric patients who have been given streptomycin. Five patients with acute renal failure who had received 1.5 to 10 gm. of streptomycin were treated by hemodialysis as early as was possible (3 to 9 days). None developed signs of toxicity. Accumulating experience emphasizes the importance of estimating renal function before giving a course of streptomycin (after authors' summary).

E. A. Riley

Value of Achromycin® (Tetracycline) in the Treatment of Pulmonary Tuberculosis (in Turkish). N. Menemenli.* *Tuberk. Toraks*, January-April, 1959, 7: 93–99. (*U. of Ankara, Turkey)

Tetracycline (Achromycin®) was used for the treatment of 33 cases of pulmonary tuberculosis, together with streptomycin and isoniazid. No particular change which could be attributed to the tetracycline was observed in the response to treatment. Susceptibility studies revealed that tetracycline delays the resistance to streptomycin and/or isoniazid; therefore, this drug may be used in lieu of PAS when for some reason PAS cannot be given.

S. Solu

Results of Specific Treatment of Miliary Tuberculosis in Children: A Follow-up Study of 63 Patients Treated with Antimicrobial Agents. E. M. Lincoln* and F. Hould. *New England J. Med.*, July 16, 1959, 261: 113–120. (*Bellevue Hosp., N. Y., N. Y.)

The prognoses of 102 children with acute generalized miliary tuberculosis seen from 1926 to June, 1944, are contrasted with results in 63 patients treated during the following 12 years with antimicrobial therapy. A small group of 9 children were first treated with a sulfone, thiazolsulfone. Six of the 9 survived and remained well until 2 of them manifested renal tuberculosis and 1, pulmonary tuberculosis 11 or 12 years after the original diagnosis of miliary tuberculosis. The case fatality rate improved in the next group of 29 children treated with added streptomycin, but meningitis developed in 6 patients under treatment for miliary tuberculosis. In the third group of 25 children treated with the addition of isoniazid, only 1 death occurred, and meningitis did not develop under treatment. Two children had tubercle bacilli recovered on culture from the cerebrospinal fluid, and the possible diagnoses of serous meningitis and abortive caseous meningitis are discussed.

As better antimicrobial agents were used in treatment, the amount of calcification seen both in the primary complex and in metastatic areas decreased. The rate of roentgenographic clearing was slowest in the cases treated by thiazolsulfone only—32 to 44 weeks; in the two later groups, with added streptomycin in one group and with three antimicrobial agents including isoniazid in the final group, it was an average of 13 weeks (after authors' summary).

M. J. Small

Clinical Course of Pulmonary Tuberculosis due to Isoniazid-resistant Bacilli (in French). J. Viallier,* C. Ollagnier, and L. F. Perrin. *Rev. tuberc., Paris*, February-March, 1959, 23: 287–290. (*Lyon, France)

The course of tuberculosis was studied in 44 patients who had previous chemotherapy which included isoniazid, and who were admitted to the hospital with isoniazid-resistant bacilli. The level of resistance was mostly over 20 γ. Catalase activity was nil or diminished in most cases. Whenever virulence for guinea pigs was studied, it was found to be diminished or absent. Among the 44 patients, 9 improved, 12 remained stationary in a very serious and probably incurable condition, 20 showed marked worsening with very poor response, and 2 died of tuberculosis. Thus, there is no correlation between low virulence for the guinea pig and the course of tuberculosis in man. While in some cases isoniazid resistance could be attributed to faulty conduct of chemotherapy, in most cases it had been correctly administered, including various drug combinations. The appearance of isoniazid resistance in these cases is attributed to initial anatomic conditions and preexisting endobronchial lesions.

V. Leites

PULMONARY PHYSIOLOGY

Effects of Alternating Positive and Negative Endotracheal Pressures on the Caliber of Bronchi. S. Lynch,* A. Levy, and K. Ellis. *Anesthesiology*, May-June, 1959, 20: 325–329. (*Columbia U., N. Y., N. Y.)

The purpose of this study was to determine, under controlled conditions in human patients, the effects of alternating positive and negative airway pressures on the caliber of the bronchi. The method of study used 5 healthy female patients under thiopental, nitrous oxide-oxygen anesthesia. Propyliodine (Dionosil Oily®) is introduced through a plastic catheter via the endotracheal tube. Roentgen films are taken with a Fairchild roentgenographic camera while simultaneous airway pressures are recorded by a spe-

cially designed radiopaque aneroid manometer. A significant reduction in the caliber of intralobular bronchi is noted in all cases. The degree of negative pressure reflects the degree of narrowing of the bronchi. This narrowing of the bronchi during the negative phase is explained as the cause of audible wheeze so commonly heard in patients under general anesthesia in their phase of exhalation. The study suggests reasons for the reduction in pulmonary complications in surgery cases where negative pressure application has been used via the airway.

E. SHABART

The Mechanical Work of Breathing during Muscular Exercise in Patients with Cardiorespiratory Insufficiency (in Italian). G. MILIC EMILI,* J. M. PETIT, and R. KOCH. *Med. lavoro*, February, 1959, 50: 108–113. (*U. of Nancy, France)

The oxygen uptake and the work of breathing were measured in 9 patients, 40 to 60 years old, with cardiorespiratory insufficiency, and in 3 normal persons, 27 to 31 years old, during muscular exercise of different intensity. The work of breathing was determined by taking the endoesophageal pressure and calculating the volume of the breathed air using Fleische's metabograph.

From the results obtained in this study, it appeared that, for a given value of pulmonary ventilation or of oxygen uptake, the mechanical work of breathing was higher in the patients than in the normal persons, and that the difference increased according to the intensity of the work performed.

I. ARCHETTI

The Effect of Changes in Ventilation and Pulmonary Blood Flow on the Diffusing Capacity of the Lung. G. M. TURINO,* M. BRANDFONBRENER, and A. P. FISHMAN. *J. Clin. Invest.*, July, 1959, 38: 1186–1201. (*Presbyterian Hosp., N. Y., N. Y.)

The effect of minute ventilation and pulmonary blood flow on the diffusing capacity of the lung for carbon monoxide was investigated at rest and during exercise by the steady-state methods. For this purpose, normal subjects were contrasted with patients in whom pulmonary blood flow had been restricted by mitral stenosis. To vary ventilation and blood flow independently, special methods, such as voluntary hyperpnea and unilateral occlusion of a pulmonary artery, were also employed. The results indicate that the diffusing capacity for carbon monoxide is little affected by changes in pulmonary blood flow until the flow is markedly reduced. By way of contrast, increases in ventilation are associated with increases in diffusing capacity, and seem to account for the rise in diffusing capacity observed during moderate exercise.

E. DUNNER

Studies on the Role of Intermittent Inspiratory Positive Pressure Oxygen Breathing in the Treatment of Pulmonary Edema. W. F. MILLER* and B. SPROULE. *Dis. Chest*, May, 1959, 35: 469–479. (*U. of Texas, Austin)

The patient with pulmonary edema is ill in direct proportion to the magnitude of the physiologic alterations. This condition frequently presents with great urgency so that immediate specific therapy is demanded. The present report demonstrates and discusses the physiologic basis for the use of oxygen administered by intermittent positive pressure breathing to more than 550 patients in the past 4 years.

Pulmonary edema, regardless of the etiology, is characterized by severe dyspnea owing to acute hypoxia and profound increase in the work of breathing with acute ventilatory insufficiency. Not infrequently, hypotension and shock are present. Oxygen and bronchodilator aerosol administered by inspiratory positive pressure breathing, with intermittent anti-foaming agents, have been demonstrated to be a safe, rapid method of correcting the physiologic disturbances, including shock. This means of therapy: (*1*) improves ventilation to relieve hypoxia and increase lymphatic flow; (*2*) relieves dyspnea by decreasing the work of breathing; (*3*) decreases transcapillary pressure gradient, and (*4*) decreases right heart filling.

E. A. ROUFF

A Physiologic Approach to the Diagnosis of Acute Pulmonary Embolism. E. D. ROBIN,* D. G. JULIAN, D. M. TRAVIS, and C. H. CRUMP. *New England J. Med.*, March 19, 1959, 260: 586–591. (*Harvard Med. School, Boston, Mass.)

In the normal subject, peak end-tidal carbon dioxide tension represents alveolar carbon dioxide tension, and is essentially equal to arterial carbon dioxide tension. This equality is based on the fact that carbon dioxide equilibration between alveolar air and pulmonary capillary blood is complete.

Carbon dioxide tensions in expired air were measured continuously by means of a rapid-acting Liston-Becker infrared carbon dioxide analyzer. Arterial carbon dioxide content was measured in a Van Slyke apparatus. A total of 24 normal subjects and 11 patients with a suspected diagnosis of pulmonary vascular obstruction were studied. In the normal subjects the mean difference between arterial and end-tidal carbon dioxide tensions was 0.1 mm. The greatest discrepancy be-

tween arterial blood and alveolar air in any one normal subject was 5.3 mm. of mercury.

In 8 of 11 suspected cases of pulmonary embolism, significant differences between arterial blood and end-tidal carbon dioxide tension were found. In 4 of these patients, conclusive evidence of pulmonary artery obstruction was obtained at post-mortem examination, surgery, or roentgenographic study. In the other 4 patients the clinical evidence was consistent with the results of the technique. In 3 patients, no significant difference between arterial and end-tidal carbon dioxide was found. In 1 of these, no pulmonary emboli were found at post-mortem examination. No definitive diagnosis was made in the other 2 patients.

Since a standard deviation for end-tidal versus arterial carbon dioxide tensions of 1.8 mm. of mercury is found in normal subjects, and there is an error in arterial carbon dioxide tension of ±2 mm. of mercury, a significant difference between the two measurements would be no less than 5 mm. of mercury. Thus, small pulmonary emboli cannot be diagnosed by the present method. This fact sharply limits the applicability of the technique since many pulmonary emboli are small.

When pulmonary infarction occurs as a result of embolism, alveoli are occluded and, therefore, ventilation as well as perfusion is decreased in the affected part of the lung. With a decrease in both ventilation and perfusion, any difference between end-tidal and arterial carbon dioxide will be decreased. Since infarction tends to take place at a later stage than occlusion, the earliest possible use of the test is desirable.

The major limitation of this procedure is that a difference in end-tidal and arterial carbon dioxide tensions is not specific for pulmonary embolism. Any lung disease associated with major changes in ventilation-perfusion relations will give rise to significant differences between arterial and end-tidal carbon dioxide tension. The most important lung disease in this respect is obstructive emphysema.

In the patient with suspected pulmonary embolism, simultaneous measurement of arterial and end-tidal carbon dioxide tensions provides useful information regarding the possibility of this diagnosis. In the absence of pulmonary infarction, a difference of less than 5 mm. of mercury implies either no embolism or an embolus occluding less than the equivalent of a lobar branch of the pulmonary artery. In the absence of emphysema, a large difference indicates pulmonary vascular occlusion. Like most tests in clinical medicine, this method must be used in association with other findings, and the limits of error recognized. The diagnostic accuracy of this test will probably improve as more accurate methods become available for measuring end-tidal and arterial carbon dioxide tensions.

M. J. Small

Pulmonary Function in the Normal Male. E. L. Holmes.* *J. Appl. Physiol.*, July, 1959, 14: 493–495. (*Wayne State U. College of Med., Detroit, Mich.)

Selected pulmonary function tests, total vital capacity (TVC), 1.0-second expiratory capacity (EC), and total expiratory time, were given to 130 healthy males aged 20 to 25 years. The mean TVC as measured in the sitting position was 5.09 l. ± S.E. 0.05. The mean value was significantly higher than predicted, using the formula of West. TVC increased with increasing body surface and height. Especially good correlation was seen between TVC and essential body mass (EBM) when an essentially linear relationship was obtained. Data from this study on a narrow age group indicate that the normal may deviate from the mean by as much as 20 per cent. The mean 1.0-second EC was 90.8 per cent ± S.E. 0.44 of the TVC. This was somewhat higher than has been reported. The total expiratory time was 1.63 seconds S.E. 0.03. There was no correlation of total expiratory time with any body measurement (after author's summary).

A. L. L. Bell, Jr.

Normal Distribution of Pulmonary Function Between the Lungs. V. Autio.* *Ann. med. int. Fenniae*, Fasc. 1, 1959, 48: 9–13. (*Paimio Sanatorium, Preitilä, Finland)

Bronchospirometric studies were performed on 22 healthy volunteers to determine the normal ratio of the functions of both lungs and its physiologic variation. The results estimated the practical mean value of the normal ratio to be 53:47 (right to left lung).

E. Dunner

A Study by Cardiac Catheterization of Pulmonary Hemodynamics in Silicotic Patients (in Italian). M. Magistretti,* E. Sartorelli, and E. Perrone. *Med. lavoro*, January, 1959, 50: 45–56. (*U. of Milan, Italy)

Pulmonary hemodynamics were studied by means of cardiac catheterization in 30 patients with different forms of silicosis. In most of the cases an increase of the total vascular pulmonary resistance (particularly the arterial) was found.

Two main factors should be considered in the pathogenesis of these modifications: (*1*) an anatomic factor, due to reduction of the pulmonary capillary bed and to arteriolar lesions; and (*2*) a functional factor due either to decrease of the alveolar pO_2 or to arterial anoxemia. According to

the results obtained in this study, the anatomic factor appears to be the more important in causing pulmonary hypertension in silicosis.

I. ARCHETTI

Estimation of Pulmonary Resistance by Repetitive Interruption of Airflow. J. A. CLEMENTS,* J. T. SHARP, R. P. JOHNSON, and J. O. ELAM. *J. Clin. Invest.*, July, 1959, 38: 1262–1270. (*Army Chemical Center, Maryland)

A simple technique for estimation of lung and airway resistance has been developed using repetitive interruption of air flow and nonlinear flowmetering. The theory of the method has been briefly explored with special attention to the influence of the thorax and effect of the upper airway properties. The physical properties of the thorax cause slight overestimation of resistance, and this effect is increased at the extremes of lung volume. The effect of the upper airway properties is to cause underestimation of resistance: This effect is accentuated by increased distensibility of the upper airways, especially when pulmonary resistance is high. It is concluded from resistance measurements on human subjects and from studies on respiratory analogues that the inherent error of the method ranges from about +0.2 cm. H_2O per liter per second in the normal to about −1.5 cm. H_2O per liter per second at 15 cm. H_2O per liter per second. It is pointed out that the method may develop larger errors in nonhomogeneous obstructive pulmonary disease.

E. DUNNER

Carbon Dioxide and Oxygen Blood Levels with a Carbon Dioxide Controlled Artificial Respirator. M. FRUMIN,* N. BERGMAN, and D. HOLADAY. *Anesthesiology*, May-June, 1959, 20: 313–320. (*Columbia U., N. Y., N. Y.)

The purpose of this study was to explore the efficiency of a mechanical respirator carrying out gas exchange in artificial respiration when compared with spontaneous breathing. The precise control of inspiratory oxygen and the end-tidal carbon dioxide tension provides the tool to study different facets of artificial respiration. The method, employing 50 healthy adults, anesthetizes and ventilates artificially with exact nitrous oxide and oxygen mixtures by use of a carbon dioxide servo-controlled respirator. Correlation studies of arterial and end-tidal PCO_2 are done, as well as correlating of desired and attained arterial and end-tidal PCO_2. Variations in buffer base during the artificial respiration and anesthesia are recorded. Arterial oxygen saturation is likewise studied.

Desired carbon dioxide tensions occur in end-tidal air, but greater variability occurs in arterial blood. Arterial buffer base values fell approximately two milliequivalents per liter. Oxygenation occurs reasonably well when inspiratory oxygen concentrations are 21 to 35 per cent.

E. SHABART

Treatment of Respiratory Failure in Two Patients with Pneumonia and with Restricted Pulmonary Function. M. BECK,* E. BERGLUND, and B. O. SIMONSSON. *Acta med. scandinav.*, May 9, 1959, 163: 467–472. (*U. Lung Clinic, Rënströmska Sjukhuset, Gothenburg, Sweden)

Two patients are reported in whom pneumonic infection was superimposed upon previously diseased lungs. Severe respiratory failure developed, with CO_2 retention and hypoxemia. Improved ventilation alone did not suffice to normalize the arterial oxygen saturation. Addition of oxygen to the inspired air further improved the oxygen saturation. The treatment was guided by means of frequent blood-gas determinations. Adequate airway drainage and ventilation may be accomplished in several ways, but is often facilitated by tracheotomy.

E. DUNNER

Respiratory Studies in Children: VII. A Longitudinal Study of the Lung Volumes in Asthmatic Children during Symptom-Free Periods. S. KRAEPELIN.* *Acta paediat.*, July, 1959, 48: 335–344. (*Karolinska Institutet, Stockholm, Sweden)

The lung volumes of 18 asthmatic children (9 boys and 9 girls) with initially elevated ratios of residual volume to total capacity were re-examined during symptom-free periods after intervals of about 2 to 4 years. In these later examinations the original hyperinflation had successively diminished, and at the last examination the lung volumes did not differ markedly from those of healthy children.

The general clinical improvement observed in the children between the first and the last examinations was closely analogous to the changes in lung volumes demonstrated during the same period. The results suggest that the hyperinflation demonstrated earlier is of functional origin and, thus, reversible. The successive normalization of the lung volumes is a hopeful sign from the prognostic aspect (Author's summary).

H. J. SIMON

The Classification and Performance of Respiratory Valves. R. FOREGGER.* *Anesthesiology*, May–June, 1959, 20: 296–308. (*Marquette U., Milwaukee, Wis.)

Definitions of terms used in connection with the design of valves are given. A classification of six

types of valves determined by the principle of their operation is discussed. A detailed report is presented on the performance of respiratory valves in regard to: (*1*) resistance to gas flow, (*2*) leakage which may be dynamic or statie, (*3*) opening pressure, and (*4*) location of the valve. The importance of these four points is disclosed and a need for cooperative research among anesthesiologists, engineers, and mathematicians is pointed out.

E. SHABART

Behavior of Respiratory Function after Partial Pulmonary Exeresis in Tuberculosis (in Italian). E. CAPODAGLIO* and A. ORLANDONI. *Gior. ital. tuberc.*, January–February, 1959, 13: 22–29. (*U. of Pavia, Italy)

Respiratory function was studied in 25 patients, 20 to 47 years old, 40 to 50 days after either lobectomy or segmental resection. According to the results obtained by measuring the values of the different components, it is concluded that, after pulmonary resection, there was always a reduction of the vital capacity and an increase of the residual volume, and that the portion of the vital capacity which was useful during hyperventilation was not reduced.

The alveolar ventilation during rest was good but not uniform in comparison with the perfusion, so that the difference of partial tension of oxygen between alveolar air and arterial blood increased. During effort, the ratio of calories per liter decreased, probably due to the fact that the alveolar surface and the pulmonary capillary bed were both reduced, and due to the special conditions of circulation depending on the prolonged inactivity.

I. ARCHETTI

MICROBIOLOGY AND IMMUNOLOGY

Action of Kanamycin on the Morphology of *Mycobacterium Tuberculosis* as seen by the Electron Microscope (in Italian). V. NITTI* and R. VIRGILIO. *Arch. tisiol.*, January, 1959, 14: 3–29. (*U. of Naples, Italy)

The action of kanamycin on *M. tuberculosis* strain H37Rv grown in Tween® 80 was followed by electron microscopy. Different amounts of the drug (2–5–10–50 γ per ml.) were added to 48-hour cultures, samples were taken at intervals of 1, 2, 3, 4, 7, and 12 days, and the results were compared with the phases of the growth curve.

Two types of alterations were mainly seen: the first was morphologic, with formation of filamentous forms, an expression of an atypical process of division, which was particularly present in cultures containing low amounts of kanamycin; the second was degenerative with final lysis, and was found among the cells in contact with the highest concentration of the drug. Apparently, normal bacterial cells were always seen, even with the highest concentration of kanamycin, and in the later samples: they were probably partially protected in small clusters of cellular material.

I. ARCHETTI

Experimental Studies on the Mode of Action of Kanamycin. I. Effect of Kanamycin on Adaptive Oxidation of Benzoic Acid, Niacin, and Malonic Acid by Avian Tubercle Bacilli (in Japanese). A. HAYASHI.* *Kekkaku*, July, 1959, 34: 463–466. (*Osaka U. School of Med., Osaka, Japan)

A study was made with the Warburg manometric technique, using the Takeo strain of avian tubercle bacilli which were resistant to kanamycin or streptomycin, or susceptible to both of them and to kanamycin at 50 to 100 γ per ml. (*1*) The "adaptive oxidation (*Suda*)" of these three substrates by avian tubercle bacilli was remarkably or completely inhibited by a small amount of kanamycin. (*2*) It also inhibited the successive adaptive oxidation of benzoic acid by avian tubercle bacilli. That is, when benzoic acid was completely oxidized by the bacilli, pyrocatecol was added, which was promptly oxidized in the absence but not in the presence of kanamycin. (*3*) Adaptive oxidation of benzoic acid and niacin by streptomycin-resistant avian tubercle bacilli was not inhibited at all by a large amount of streptomycin, but it was completely inhibited by kanamycin. (*4*) The same and comparable phenomena were observed with kanamycin-resistant bacilli and with kanamycin or streptomycin, respectively.

I. TATENO

Atypical Mycobacteria and Drug-Resistant Tubercle Bacilli Isolated During a Survey of Untreated Patients with Pulmonary Tuberculosis. J. B. SELKON* and D. A. MITCHISON. *Tubercle*, June, 1959, 40: 141–154. (*Postgraduate Med. School, London, England)

During a national survey of the incidence of drug-resistant tubercle bacilli from patients with newly diagnosed untreated pulmonary tuberculosis, strains resistant to streptomycin, PAS, and isoniazid were obtained from 55 of 1,081 patients with positive cultures. These resistant strains were identified by *in vitro* and animal virulence tests to distinguish atypical strains from tubercle bacilli.

The 7 atypical strains from the survey, with an eighth obtained elsewhere, consisted of 1 strain of *M. tuberculosis* var. *avium*, 1 strain with a very close resemblance to human tubercle bacilli, 5 photochromogenic strains, and 1 scotochromogen. There was suggestive evidence that all except the scotochromogen were of pathogenic significance in the patients. Chromogenic atypical strains

could easily be distinguished by their pigmentation and high catalase activity.

The 3 *in vitro* identification tests that were found to be of greatest value in separating atypical strains from tubercle bacilli were the study of colonial morphology on oleic acid–albumin agar plates, the niacin test, and the thiosemicarbazone sensitivity tests. Slightly less satisfactory were the arylsulphatase test, the formation of cords in slide culture, and catalase activity. Bacillary morphology, the methylene-blue reduction time, and the neutral red test were of limited value. All atypical strains failed to cause progressive disease in guinea pigs, but the photochromogens were as pathogenic to mice as were tubercle bacilli. Three PAS-resistant bovine strains of tubercle bacilli had rapid methylene-blue reduction times. Procedures for the routine detection of atypical strains are suggested (after authors' summary).

M. J. Small

Studies on a Strain of "Chromogenic Pathogenic Mycobacteria" Isolated from Resected Human Lung Lesions. Bacteriologic, Pathologic, and Clinical Findings (in Japanese). Y. Miyamoto,* T. Iwasaki, K. Iwai, S. Adachi, S. Miyasaka, S. Okamoto, S. Ito, and T. Araki. *Kekkaku*, July, 1959, 34: 498–502. (*Kanagawa Prefectural Pub. Health Lab., Yokohama, Japan)

Clinical, histopathologic, and bacteriologic studies were carried out in a patient from whose resected lung lesions a strain of "chromogenic pathogenic mycobacteria" was isolated. It was shown that the virulence of this strain in guinea pigs fell between that of *M. tuberculosis* on the one hand, and nonpathogenic mycobacteria on the other, the situation closely simulating that of enteropathogenic coli groups lying between Shigella and *Escherichia coli*. Its pathogenicity for man and experimental animals appeared to depend upon some unknown factors of the host itself.

The patient was a 29-year-old female whose tuberculin reaction was "doubtful" in 1953; but the roentgenogram revealed thumb-sized cavity and infiltrations in the right middle lung field in September, and exudative pleurisy developed on the same side in November. The cavity became smaller on streptomycin and PAS therapy. Sputum culture was constantly negative, but she expectorated a creamy mass in June, 1954, which contained acid-fast bacilli. Acid-fast bacilli were demonstrated twice by fluorescent microscopy on other occasions. The tuberculin reaction was strongly positive in 1955, and surgery was done in May, 1956.

The cavity was inspissated, and its wall was fibrous and thick; it was accompanied by caseous bronchitis and showed quite the same picture as a tuberculous cavity. Only chromogenic mycobacteria were cultured in a pure form. The colony of the bacilli on culture medium was yellow-orange, wet, glistening, photochromogenic, and grew best at 32°C. Epithelioid tubercles were formed in guinea pig lung upon inoculation. They grew best in the spleen, followed by the lung; they were recovered from the lung for about 3 weeks, and from the spleen for about 4 weeks. They were susceptible to 5 γ each of streptomycin, viomycin, kanamycin, and isoniazid per ml., 100 γ of PAS per ml., and were resistant to other ordinary antimicrobials.

I. Tateno

Studies on Mycobacteriophage (in Japanese). Y. Hikawa.* *Kekkaku*, July, 1959, 34: 467–474. (*Institute of the Tuberculosis Prevention Society, Tokyo, Japan)

Mycobacteriophages were isolated from 14 soil samples (9 strains) and from 11 samples of animal or chicken feces (6 strains) which were enriched by adding suspensions of washed saprophytic acid-fast bacilli. The pH value of the material was not related to the incidence of phage isolation. One group of attenuated mycobacterial strains was lysed by many of the 15 phage strains, but the other slightly more virulent tubercle bacilli were lysed only by a part of the phage strains employed. One of the phage strains (M9) was lytic against the H37Ra and 5 strains of human tubercle bacilli, as well as 2 strains of virulent avian tubercle bacilli (Cecillie and E 38686), which was unusual because there has so far been no report of mycobacteriophage that was active against virulent avian tubercle bacilli. These findings suggest that classification of mycobacteria by phages may be possible. Tween® 80 and human sera inhibited the activity of the phage, but the presence of the 2 reactivated the mycobacteriophage.

I. Tateno

EXPERIMENTAL PATHOLOGY

Studies on Deposition of Submicronic Dust Particles in the Respiratory Tract. L. Dautrebande,* H. Beckmann, and W. Walkenhorst. *A. M. A. Arch. Indust. H.*, April, 1959, 19: 383–391. (*Silicosis Research Unit, Bochum, Germany)

An experimental study of the lung deposition characteristics of four air-borne particles less than a micron in size (India ink, calcium carbonate, iron oxide, and aluminum powder) was undertaken on 3 normal human subjects breathing at three different rates. The mean size of the India ink particles was approximately 0.1 μ and that of the other particles about 0.2 μ.

The deposition rate of these submicronic particles in the lungs taken as a whole and in the last portion of the tidal air varies inversely with the respiratory rate. The deposition rate of these submicronic particles in the deep pulmonary regions, as represented by the Haldane-Priestley alveolar air, is not influenced by the respiratory rate. The alveolar deposition of these submicronic particles was regularly found to be very high (90 per cent and more of the inhaled particles). Except for India ink (the maximal particle size of which is 0.5 μ and the deposition of which consequently takes place exclusively in the deepest pulmonary regions), the mean size of the particles collected in the various portions of the expired air was observed to decrease regularly toward the deep alveolar air.

The larger the granulometric spectrum of the inhaled particles, the greater the difference in the mean size and in the size distribution curves between inspired dust-laden air and global expired air, end tidal air, and alveolar air. The maximal size for particles collected from deep alveolar air and photographed with the electron microscope was found to be around 1 μ. None of these findings appeared to be influenced by the number of particles present in the inspired air (from 15,000 to 400,000 per cc.

T. H. NOEHREN

Clinical Syndromes Associated with Deficient Lung Fibrinolytic Activity. I. A New Concept of Hyaline-Membrane Disease. J. LIEBERMAN.* *New England J. Med.*, March 26, 1959, 260: 619–626. (*U. of California School of Med., Berkeley)

Tissue fibrinolytic activity was studied in the lungs and several other organs of 49 fetuses and infants dying perinatally, including 8 with pulmonary hyaline-membrane formation. The tissue activator of plasminogen was found to appear in the lung as early as the third month of gestation: 33 of 41 fetuses without pulmonary hyaline-membrane formation showed this activity. However, none of the 8 infants with hyaline membranes showed this activity. Guinea pigs and rabbits—animals subject to hyaline-membrane formation from oxygen poisoning—demonstrated this same deficiency, in contrast to rats, whose lungs contained the enzyme and were more resistant to formation of the membrane.

Hyaline-membrane formation was induced in guinea pigs by exposure to toxic concentrations of oxygen. This lesion appeared much more readily when oxygen poisoning was supplemented by nebulized filtered amniotic fluid. A preliminary review of the reproductive histories of mothers who delivered infants with confirmed hyaline-membrane formation shows a tendency to multiple neonatal deaths suggestive of hyaline-membrane disease.

These data suggest that a predisposing factor in hyaline-membrane formation of the newborn is the absence of plasminogen-activator activity in the lung. An interaction between amniotic fluid or tissue thromboplastin and other clotting factors in an alveolar effusion results in deposits of fibrin that are transformed into "hyaline membrane" postnatally. Such fibrin deposits would be lysed normally through action of the fibrinolytic system if the tissue activator were present. The defect does not appear to be necessarily related to the degree of prematurity, but may represent a genetic aberration (after author's summary).

M. J. SMALL

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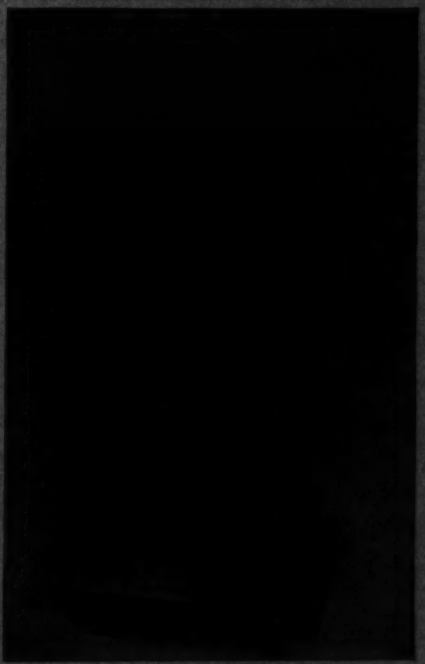

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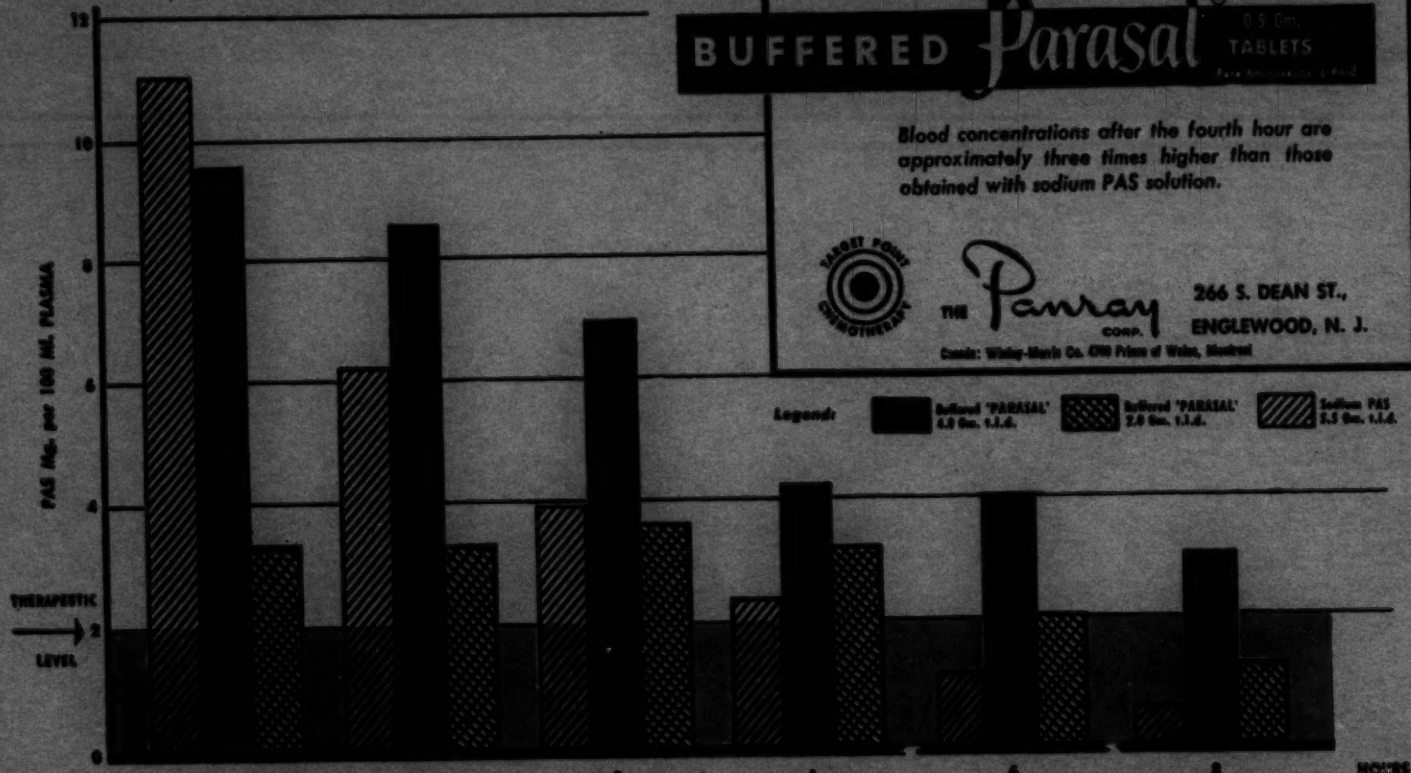

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Ref.: Doub, E. N. & Vitagliano, G.R.: A. Rev. Tuber. & Pul. Dis.: 72,543-7 (Oct. '55)

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